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Three-Dimensional Visualization of Contact Networks in Granular Material

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Three-dimensional visualization of contact networks in granular material

The research in this thesis is dedicated to the development of fluorescent dyes for quantitative imaging of contact forces in soft matter. The ultimate goal is the direct visualization of force networks in three-dimensional granular systems, in particular of the magnitude of individual contact forces between touching particles, as such providing valuable information on how forces are distributed throughout the bulk of a granular packing.
Visualization of contacts within a granular system was performed using particles covalently functionalized with highly sensitive fluorescent probes that change their photophysical properties in response to changes in their local environment. An overview of suitable approaches is given in *chapter 2*. Exemplary of such changes in photophysical properties, which are employed in this research, are *solvatochromism* and *rigidochromism*. Solvatochromic fluorescent probe molecules exhibit a bathochromic shift of their emission maximum in media of increasing polarity. It is envisioned that exclusion of solvent from the contact points alters the polarity locally, dependent on the amount of force experienced in these points. As such, the wavelength of emission would be a measure of the amount of force present in that particular point. Rigidochromism is associated with increased fluorescence intensity with increasing medium viscosity. More force experienced by the probe on the surface of the granule is expected to lead to brighter fluorescence, for example when force is applied to the system. As such, the regions within the granular bulk experiencing increased force would be identifiable.

The core experimental part of this thesis consists of chapters 3, 4 and 5. *Chapter 3* gives an overview of the experimental techniques that are used throughout this research, with the focus on fluorescence confocal microscopy, which is the most prominent experimental technique for imaging the particles and their contacts. In *chapter 4*, the synthesis and characterization of the solvatochromic and rigidochromic probes is described, as well as the modification of those probes with an appropriate linker to enable immobilization on various surfaces. The carboxylic acid functionalized derivatives of a perylene monoimide solvatochromic probe and a dicyanomethylene-dihydrofuran rigidochromic probe were synthesized in 42 % total yield over five steps and in 19 % total yield over four steps, respectively. The modifications of the molecular structure by attaching the linker turned out to have virtually no effect on the solvatochromic and rigidochromic behavior of the fluorescent probes. The emission and excitation spectra of the solvatochromic probe were studied under high liquid pressure. At higher pressures the solvent is more polar, but the orientation

contribution to the solvation decreases, resulting in a decreasing Stokes shift. The rigidochromic fluorophore exhibited a linear relationship between the logarithm of intensity of the fluorescence and the logarithm of the pressure-controlled viscosity of the medium, in accordance with the Förster-Hoffmann equation. As such, the intensity could be used to determine the pressure applied to the system.
Covalent attachment of the modified probes also requires chemical modification of the surfaces, which is described in *chapter 5*. Surfaces of poly(methylmetacrylate) (PMMA) and glass were aminolyzed with diamines and silanized with aminosilanes respectively, to enable peptide coupling reactions with the carboxylic acid modified probes. To ensure sufficient stability towards organic solvents, the use of cross-linked PMMA was found to be necessary. Thus, PMMA particles with a diameter of 10 μm and glass cover slips were successfully functionalized with solvatochromic and rigidochromic probes. Increasing the aminolysis time of the PMMA surface gave increased functionalization and thus a more intense fluorescence. Atomic Force Microscopy studies showed that the surface of the PMMA particle was significantly smoothened after functionalization with the solvatochromic probe. The rigidochromic behavior of the probe was translated to the cover slip. The immobilized probes experienced the highest viscosity when the cover slip was dried, while wetting with DMSO dramatically reduced the fluorescence intensity. When dried, the rotation in the excited state is blocked, resulting in extended fluorescence lifetimes with respect to the wetted cover slip.

In chapters 6 and 7 the results obtained with the functionalized surfaces are discussed. *Chapter 6* describes the confocal microscopy measurements on the particles functionalized at their surfaces with solvatochromic probe. The dependency of the fluorescence lifetimes, emission maxima and fluorescence intensities on the application of force was investigated for both the contact points (i.e. where the particles touch) and the non-contact points. The major goal of these measurements was to determine the extent of force sensitivity of the immobilized probe. Unfortunately, even in measurements in which the force was most directly transferred to the contact points, i.e. the single layer of particles on top of a cover slip, no variations in the photophysical properties of the solvatochromic probe were observed, which led to the conclusion that the solvatochromic probe is not directly suitable for the visualization of forces. However, as the contact points in cross sections of the particles exhibited brighter luminescence because at these points two monolayers of fluorophores are in the detected volume, the location of the contact points could clearly be visualized. As a result, the entire contact network between the particles can be reconstructed accurately.
Chapter 7 investigates the application of force on surfaces functionalized with rigidochromic probes. The application of force did not result in a variation of the fluorescence lifetime and the wavelength of emission, when a force was applied to the

system. When contact points were observed where particles touch sideways, no effect of force (applied from above) on the fluorescence intensities was detectable. On the other hand, when the bottoms of particles resting on the glass surface were observed, the fluorescence intensity significantly increased upon application of force, and was observed to vary in different contact points, suggesting that different contact points experience the applied force to a different extent. However, as no variation in the fluorescence lifetime was observed, the variation in the fluorescence intensity may have been caused to some extent by slight out-of-focus confocal imaging. In addition, it could be concluded that the probe experiences a relatively rigid environment on the PMMA surface when no force is applied, in view of the long fluorescence lifetime. Similar measurements, in which we pressed a glass bead against a functionalized cover slip showed an increasing area of the contact with increasing force, in very good agreement with continuum elasticity predictions of the elastic deformation of the materials (Hertz theory).

Chapter 8 describes the development of computer software capable of reconstructing the contact network in a system of spherical particles using PMMA particles labeled with the solvatochromic fluorophore. With such a digitally reconstructed contact network, various statistical analyses can be performed, which allow investigation of the mechanical stability of the granular system. As an example the packing structures, contact numbers and angles distribution and dependency on the z-coordinate are described. At the bottom of the system, where the glass slide forms a fixed barrier, the particles tend to be regularly packed. Higher up in the sample the order is lost.

The solvatochromic PMMA particles were used successfully for the visualization of a contact network in a three-dimensional granular system. The results in *chapter 7* indicate that the rigidochromic PMMA particles can show increased fluorescence intensity when a force is applied to the granules. We hope that after further optimization of the rigidochromic particles, such as the use of longer linkers to decrease the rigidity on the surface, they may form the first granular system in which the three-dimensional visualization of force networks is possible.