

Pharmaceutical Drug Carriers – Study by Neutrons, SAXS and DLS

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Application simulation and structure investigation : DLS + SANS

- Specific target **Nanoparticles for therapy** of cancer and other diseases were assembled from lipids, polymers, and pharmaceutical drugs or mRNA. For cell targeting proteins were bound to the surface (corona). The structure in solution is analyzed by dynamic light scattering **DLS** and **SAXS** combined with neutron small angle scattering **SANS**, SAXS, metal specific X-ray scattering **ASAXS**. Material sub-domains in the nanoscaled drug carriers (~100 nm) were localized by **Deuterium-contrast variation in SANS** and by **ASAXS**. [7, 2-4] of nanoscaled drug carriers (liposomes, solid lipid particles, micelles, magnetic oxide, and polymer-protein particles).
- The power of D-contrast SANS is the **specific detection of material domains** with different hydrogen content, e.g. of **drug, mRNA, lipid, polymer, protein**.
- The **mixed nanoparticles** (100 nm), e.g. biodegradable polymer (PLGA), protein, carbohydrates), intestinal lipid-bile nanoparticles, lipid particles, surface-proteins and optional bio-target domain are amphiphilic and partly charged. Thus the internal particle structure forms **sub-domains of different material and scattering power**, enabling a localization by contrast. For several medical cases we construct and study pharma nanoparticles for parenteral and oral applications, which contain soluble or hydrophobic drugs, or nucleic acid drugs, e.g. **mRNA** for immunotherapy of cancer and vaccination.

Therapeutic Nanoparticles as Drug carriers: Mixed nanoparticles bearing material domains



Pharmaceutical drug application:

cancer therapy, vaccination

parenteral (injection) :

study of formulations and interactions

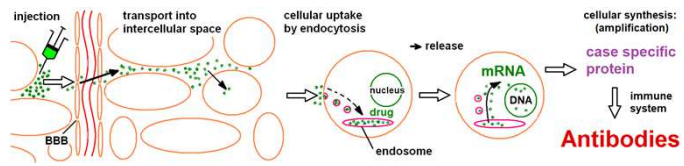


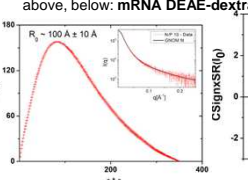
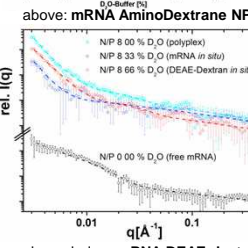
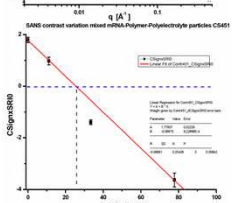
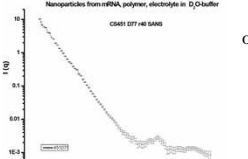
Fig.1: Drug nanoparticles for parenteral application (injection): The drug (pharma agent, mRNA) is entrapped in lipid, liposomes or polymer nanoparticles. After transfer to the relevant body region the particles are taken up into cells and release the drug, **if the structure is sufficient**. Structure and drug loading is investigated by D-contrast variation neutron small angle scattering **SANS** and **DLS**, structure details by high flux **SAXS**. In case of mRNA the active agent (protein) is formed by local bio-synthesis.

- **Parenteral nano-drug application (injection): mRNA nano-complexes for immune-vaccination and cancer therapy** [15-18] work by cellular synthesis of the corresponding case specific protein (not the antigen, but the genetic information for it is supplied). The cells and immune system of the patient work as biological drug amplifier.
- mRNA and synthetic pharmaceutical drugs can be applied in mixed nanoparticles bearing a molecular organization (domains of drug, excipients, carrier, surface ligands). Two classes cover the main drug nanoparticle forms: polymer-drug nanoparticles and liposome-lipid-drug nanoparticles. For therapeutic mRNA both classes were investigated by d-contrast SANS and the FMR2 reactor of the MLZ (KWS2 instrument), by SAXS at the DESY PETRA III (P11 instrument) and projecting **DLS** (backscattering NIBS at 170°). At the MLZ, all samples were investigated by projecting **DLS** in the SANS cuvettes (1 mm Q), immediately after the neutron study. The **SAXS** investigation at high flux revealed highly resolved substructures, while the **D-contrast SANS** studies yielded differentiation and identification of domains.

mRNA-Nanoparticles for immuno-therapy : two forms

mRNA polymer-nanoparticles:

- mRNA (protein/antibody coding)
- + structure polymer excipient
- + charged poly-electrolyte



mRNA lipid-layer nanoparticles:

- mRNA (protein/antibody coding)
- + structure lipid excipient
- + charged lipid excipient

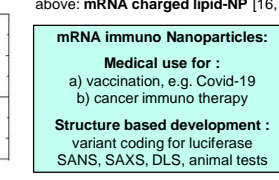
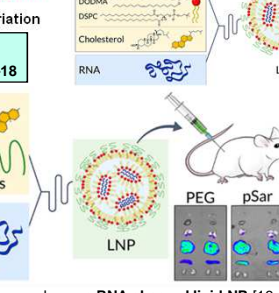
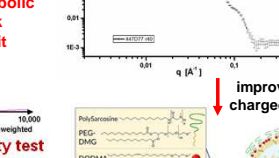
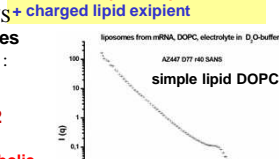


Fig.2-5: D-contrast SANS of mRNA nanoparticles for immuno-therapy :

MLZ FRM-II, KWS2
DESY-Petra III, P12

embolic risk limit
DLS as med. security test

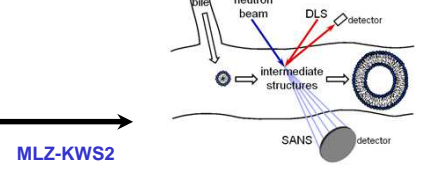
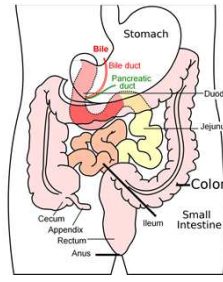
Deuterium-contrast variation
for details: see reference 15-18

Lipids
RNA

above: mRNA AminoDextrane NP
below: mRNA DEAE-dextrane NP [15, 18]

mRNA immuno Nanoparticles:
Medical use for :
 a) vaccination, e.g. Covid-19
 b) cancer immuno therapy
Structure based development :
 variant coding for luciferase
SANS, SAXS, DLS, animal tests

oral (tablet, capsule) : study in a simulator model (GI-Sim)



MLZ-KWS2
ILL-D11
for details: see contributions 239, 241

DLS: large particles 0.1 - 300 µm
SANS: nano particles 10 - 200 nm

observation by time-resolved SANS + DLS

- **Oral nano-drug application** (tablets, capsules) is tested with a simulator device of the gastro-intestinal tract with **SANS+DLS** observation of drug nanoparticles and intermediates. The structure optimized drug-carriers shall improve the application of **hydrophobic and difficult drugs** (BCS-classes 2-4), food-drug interaction problems and side effects. A specific development was the introduction of a **cholesterol-containing medium FASSIF-C**.
- At the MLZ (KWS2) and ILL (D11), all samples were investigated by **projecting DLS** (backscattering NIBS at 170°) in the SANS cuvettes (1 mm Q), immediately after the neutron study. The projecting **DLS** system contains two separate optical benches in a common carrier shining on the same focus, 120 mm in front of the device, with sufficient space for the SANS.
- **Time dependent processes** in the gastro-intestinal fluid system were studied by time resolved tr-(SANS+DLS) with a) stopped flow mixing and static sample (MLZ KWS2 and ILL D11); and b) with a constant flow through channel cuvette (200x12x1 mm Q) as position-time resolved xtr-SANS+DLS with a SM-drive (KWS2 at MLZ).

Targeting drug excipients for nontherapy

- **Tissue and cell targeting:** Intestinal, cell or tumor recognition and uptake of the drug carriers can be triggered by a surface protein or ligand head (see method sub-page targeting).

polymer PLGA-cholesterol-lipid particles for parenteral, oral, pulmonary and radio-therapy

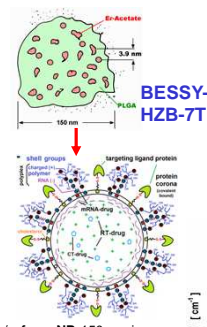
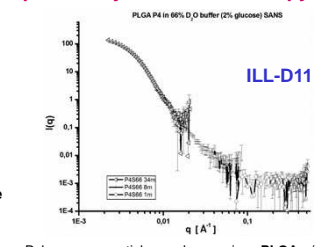


Fig.6: Therapeutic polymer nanoparticle for parenteral and oral therapy are investigated by **SANS + DLS**. For tissue and cell targeting they can be coated by a specific protein shell (cell recognition).

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