Silica nanoparticles as a carrier for virus like particles

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Introduction. Virus like particles (VLP) that consist of proteins forming outer shell of the virus are in use as travellers through blood vessels to deliver a medicine filled in the particle to the specific cells. VLP containing the medicine finds the target cells and releases the medicine with the aim of treatment. The efficiency of the treatment enhances when the concentration of VLP increases near the target cells. However, high concentration of VLP in a human body could lead to toxic effects. To avoid this, the high concentration of VLP could be supplied just in the vicinity of the target cells. To achieve this, VLP should be packed in a nanolorry that delivers them to the cell. The role of the nanolorry could be provided by a nanoparticle that is able to attach several VLP. The electrical charge is localized at the surface of VLP [1]; therefore the latter could be attached to the nanoparticle due to the Coulomb interaction. The nanoparticle must demonstrate the ability for polarization. Besides, the nanoparticle must be harmless in respect to human organism. The above conditions are met by the SiO_2 nanoparticles [2, 3] and therefore they could be in use as the nanolorry.

The goal of the work is to verify a capability of the SiO_2 nanoparticles to adhere to VLP, the hepatitis B viral capsids being in use as the model.

Methods. To study the capability of the SiO_2 nanoparticles to attach VLP the optical absorbance spectra of the VLP, SiO_2 nanoparticles and VLP mixed with SiO_2 nanoparticles (**SiO_2+VLP)** in buffered solutions were measured and compared. The Thermo Spectronic Heλios Gamma spectrophotometer that supplied wavelengths in a range 200 - 1090 nm was in use to detect absorbance.

To verify the SiO_2+VLP coupling transmission electron microscopy (TEM) and fluorescence microscopy (FM) were employed. JEOL JEM-1200EX microscope was in use for TEM, and Leica DMI 3000 B microscope for FM.

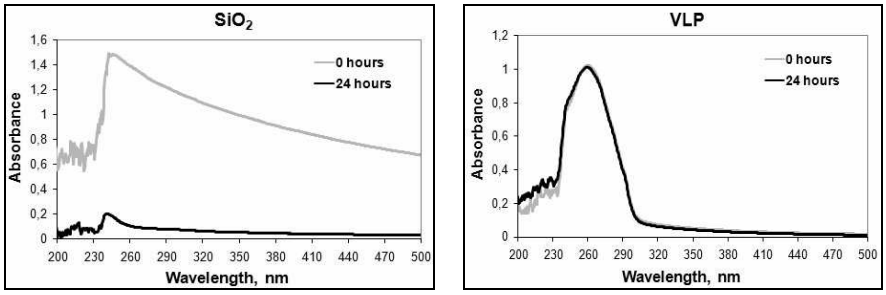
To study the fluorescence, VLP were marked with green FITC agent that forms covalent bonds with VLP amino acids. Fluorescence images were recorded with the I3 set of filters, fluorescence was excited with the 450-490 nm bandpass filter, the detection of fluorescence was done with the above 515 nm bandpass filter.

Materials. The VLP (hepatitis B viral capsids) were provided by the Biomedical Research and Study Centre of Latvia.

The SiO_2 nanoparticles were bought from the Sigma-Aldrich company. The size of the nanoparticles was 10 - 20 nm according to the Sigma-Aldrich certificate.

The buffer solution was prepared from 20 mM Tris-HCl pH 7.8, 5 mM EDTA, 150 mM NaCl, and 1 litre distilled water. NaCl had the same concentration as in the physiological solution. The concentrations of the SiO_2 nanoparticles in the buffered solutions were equal to 1 mg/ml. The concentrations of VLP were taken according to the optical absorbance (A) measurements at 260 nm, following the calibration procedure in advance. To measure A, the spectrophotometer that was mentioned above was in use. The values of A were equal to 1 (± 5%).

Results and discussion. The absorbance spectra of the solutions were tested on time stability before starting measurements. Fig. 1a demonstrates that absorbance of the SiO_2 nanoparticles solution had a drift in time (during 24 hours).



(a) (b)

Fig. 1. Drift of the SiO_2 nanoparticles and VLP solutions absorbance spectra in time: a) SiO_2, b) VLP

This could be due to the gravitation forces that move the nanoparticles towards the bottom of the measurement cell that contains the solution, a precipitation of the SiO_2 nanoparticles was observed at the bottom of the cell.

The spectra of the VLP solution did not demonstrate absorbance changes in time (Fig. 1b), no precipitation of the VLP at the cell bottom was observed. This means that VLP remain in suspended state in the solution at least during the time of the experiment.

If SiO_2 nanoparticles attach VLP, the SiO_2+VLP aggregates might fall to the bottom of the measurement cell. As the result, the concentration of VLP in the upper area of the cell decreases in time. The decrease of the absorbance in the upper illuminated part of the cell is the index of such the process.

Therefore, the changes of VLP absorbance in time were measured for the SiO_2+VLP solution to recognize a capability of SiO_2 to adhere to VLP.

The spectra of the SiO_2+VLP mixture solution demonstrate that the spectra of the VLP solution are influenced by time (Fig. 2). The absorbance decreased in time, the peculiarities (location of the maximums and bending points) of the spectra were not changed.

It evidences from the above in favour of VLP being attached to the SiO_2 nanoparticles, the physical coupling was preferable to the chemical one not to disturb the chemical bonds of VLP.

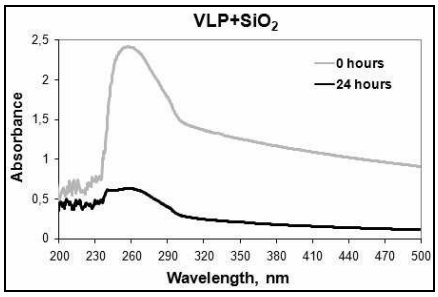
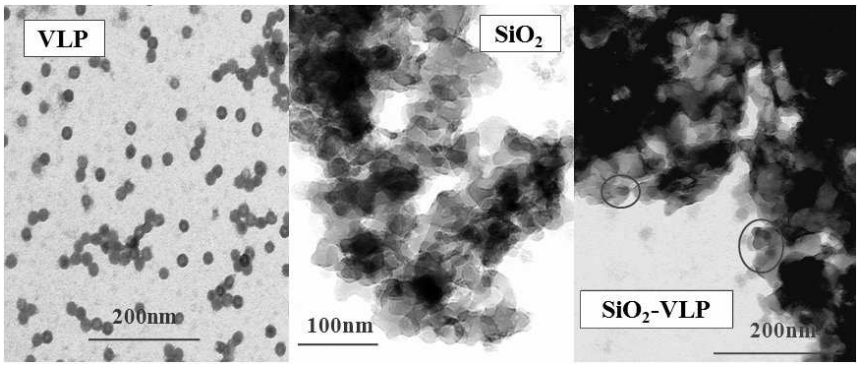


Fig. 2. Drift of the SiO_2 +VLP solution absorbance spectra in time

The electron microscopy provided the evidences on SiO_2 and VLP coupling. Fig. 3 demonstrates that the VLP (black and grey circles at Fig. 3a and 3c) were attached to the SiO_2 (Fig. 3c), the latter was imaged as the sharper black and grey particles (Fig. 3b and 3c).



(a) (b) (c)

Fig. 3. Transmission electron microscopy images of the particles:
a)-VLP, b)-SiO_2, c) SiO_2-VLP

Fluorescence microscopy gave one more evidence on SiO_2 + VLP coupling. The fluorescent VLP were observed as they connected to SiO_2 nanoparticles (Fig. 4).

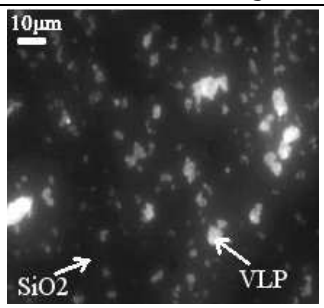


Fig. 4. Fluorescence microscopy images of SiO_2-VLP

Conclusions

1. The SiO_2 nanoparticles attached VLP by the physical coupling.
2. The SiO_2 nanoparticles could be in use to collect VLP (particularly the Hepatitis B) and perhaps deliver them to the targeted cells.

References

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By packing of therapeutic molecules, virus like particles (VLP) could be used as potential carriers of medicine for treatment of the specific cells. To increase local VLP concentration several VLP could be attached to the nanoparticle, and that is expected to increase efficiency of the treatment. It is demonstrated that SiO_2 nanoparticles attach VLP (particularly the Hepatitis B viral capsids being as the model) and perhaps could be in use as the nanolorry for VLP to increase their concentration in the vicinity of the specific cells.