Quantification study of a UV-active block copolymer and its self-organized structures

Sandro Baumgartner (Gymnasium St. Klemens, LU), Cristina Zivko (Liceo Bellinzona, TI)

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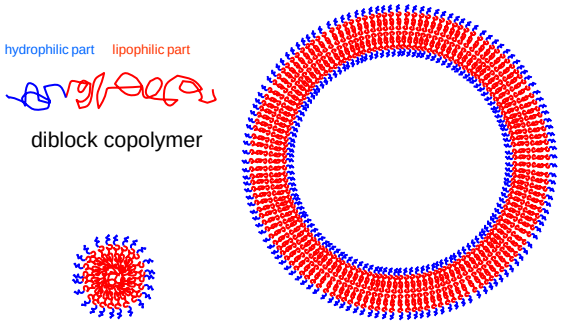
Introduction:

Polymers having several properties find increasing application in life science and material science. For our experiment we used a diblock copolymer, which consists of a hydrophilic part and a hydrophobic part, so that in a solution with water they form vesicles and micelles (scheme on the right side). These vesicles could be used for transporting hydrophilic substances, such as drugs, in human body. So far there exists no simple and accurate method to determine the concentration of such polymer vesicles in aqueous solution.

Our project consisted on producing vesicles out of the amphiphilic poly(dimethylsiloxane)-*b*-poly(2-methyloxazoline) PDMS-*b*-PMOXA block copolymers having a UV-acitve molecule on each end. We developed a method to determine their concentration by UV-spectroscopy. Therefore we calculated the extinction coefficient of the UV-active diblock copolymer at its absorption maximum in tetrahydrofuran.

We lyophilized a defined volume of aqueous vesicle solution. Afterwards we solved the residual polymer in a defined volume of tetrahydrofuran and measured its concentration by UV-spectroscopy. From the measured polymer concentration in tetrahydrofuran we are able to calculate the exact polymer concentration in aqueous solution.



Material and Methods:

1. Filmrehydration & Extrusion

We solved the UV-active polymer in ethanol andevaporated it in a round bottomed flask, so that a thin film was formed. After we added water and stirred the solution over night to form vesicles. A lipex extruder (picture below) with a track etched membrane was used to reduce the average vesicle diameter to 200 nm.

2. UV-Spectroscopy measurements

Different concentrations of the UV-active polymer were measured in tetrahydrofuran at the absorption maximum (251 nm). Out of these data we calculated the extinction coefficient ε of the UV-active polymer. Therefore we used the Lambert-Beer law:

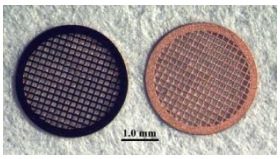
$$\log\left(\frac{I_0}{I}\right) = c * d * \varepsilon$$

Where I_0 is the intensity of the incident light beam, I is the intensity of the light beam after passing the sample, c is the concentration in mg/mL and d is the thickness of the measuring cuvette.

3. Transmission Electron Micrscopy

Transmission electron microscopy was used to image the extruded vesicles. We incubated 5 µL of the vesicle solution on a charged TEM-grid (picture below). Then we washed the sample with a uranyl acetate solution (concentration of 2%) to stain the sample.

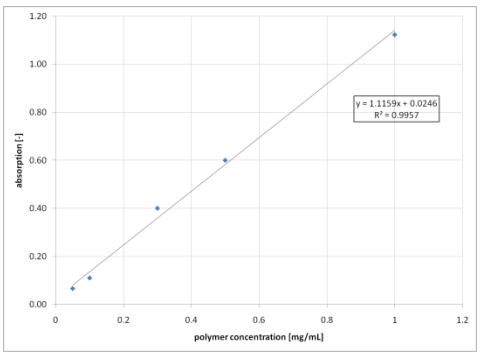
After the sample preparation we performed the TEM measurements with a Philips Morgani 268D setup with a tungsten source operated at 80 kV.

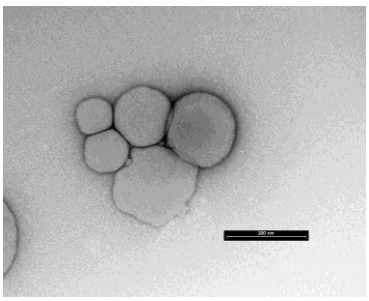


Results:

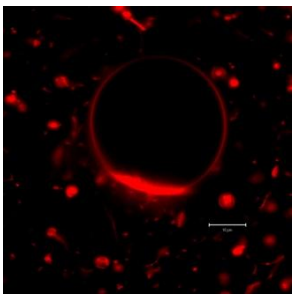
The diagram below shows the calibration curve of the UV-active diblock copolymer in tetrahydrofuran. From the linear calibration function of this polymer, we calculated the extinction coefficient of the UV-active polymer in THF. The value is ε_{251nm} = 1.14 mL/(mg*cm)

After lyophilization of 3 mL of the aqueous vesicle solution we dissolved the residue in 2 mL of THF. The measured absorption of 0.061 corresponds to a polymer concentration of 0.054 mg/mL in THF. This means that we have a diblock copolymer concentration of 0.036 mg/mL in water.





From electron transmission microscopy images we can assume that we have polymer vesicles with diameters around 200 nm in the aqueous solution. One example of such vesicles is shown in the TEM image above. The black scalebar represents 200 nm.



To prove that the polymer forms vesicles in aqueous solution, we imaged sulforhodamine B labelled polymer vesicles with a Zeiss 510 Confocal 2 microscope equipped with an argon-ion laser to excite the dye at 514 nm.
As we can see in the micrograph above, we obtained vesicles having diameters up to 40 µm. The white scalebar represents 10 µm.

Acknowledgements:

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