Methacrylamide-oligolactates as building blocks for targeted biodegradable polymeric micelles to deliver photosensitizers

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Introduction

Amphiphilic block copolymers consisting of poly(ethyleneglycol) (PEG) and p(methacrylamide-oligolactates) self-assemble in aqueous media into spherical micelles above the cloud point (CP) of the thermosensitive block [1,2]. Cleavage of the lactic acid side chains causes a gradual increase of the critical micelle temperature (CMT) and leads to controlled destabilization of the micelles (Figure 1).

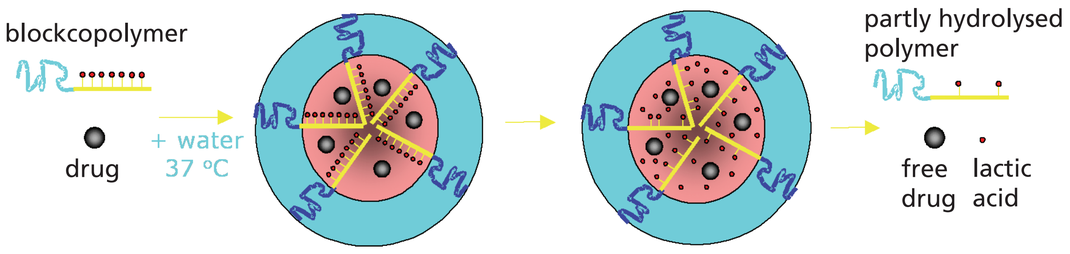


Figure 1 General concept of thermosensitive biodegradable polymeric micelles.

The hydrophobic micellar core and its destabilization at physiological conditions can be exploited for controlled delivery of hydrophobic drugs at their site of action .
The hydrophobicity of many photosensitizers, which are applied in photodynamic therapy (PDT), results in severe side effects and necessitate that they are encapsulated in an adequate carrier system to deliver them specifically to tumour regions (Figure 2).

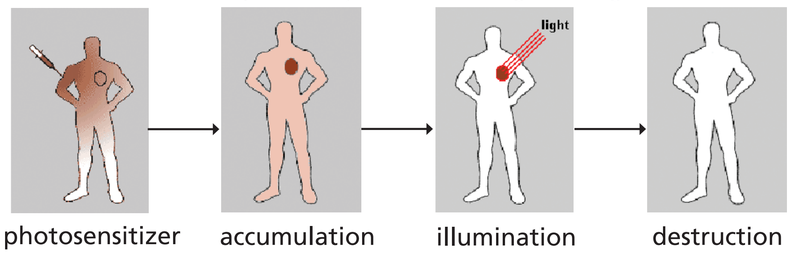


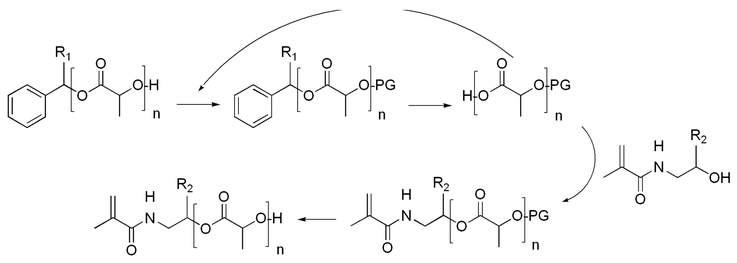
Figure 2 Photodynamic therapy.

Aim

The application of biodegradable thermosensitive polymeric micelles and their use for targeted delivery of photosensitizers to cancer cells.

Synthesis monomers and polymers

Monodisperse methacrylamide oligolactates were synthesized in a stepwise manner [1] (Scheme 1).



Scheme 1 Step by step synthesis of monodisperse methacrylamide-oligolactates. R, and R_2 are H or CH_3; PG = protecting group

Free radical polymerization of these monomers with either the macroinitiator $(CH_3\text{-O-PEG})_2$-ABCPA or $(NH_2\text{-PEG})_2$-ABCPA (ABCPA = 4,4- azobis (4-cyanopentanoic acid)) resulted in amphiphilic thermosensitive block copolymers.

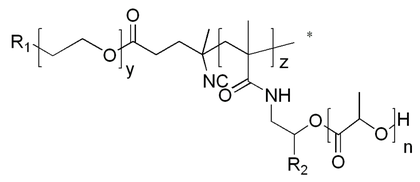
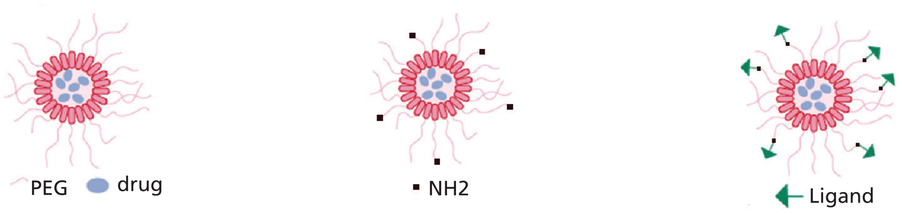


Figure 3 Chemical structure of a $R_1\text{-PEG}_{5000}$-b-p(HE/PMAm-Lac_n) block copolymer. n is 1 to 4, $R_1 = OCH_3$ or NH_2, $R_2 = H$ or CH_3.

Targeted Micelles



The primary amines on the micellar surface can be used to covalently link, via a bifunctional coupling agent, targeting ligands (e.g. antibodies).

The characteristics of some amphiphilic block copolymers are shown in Table 1.

Table 1: Characteristics of biodegradable thermosensitive polymers and their micelles based on $mPEG_{5000}$-*b*-p(HE/PMAm-Lac_n).

Polymer composition	M_n (GPC)	CMT (°C)	CMC* (mg/ml)	Z_{Ave} (nm)	Destabilisation time period (in hours; pH 7.4; 37°C)
$mPEG_{5000}$-*b*-p(HPMAm-Lac_2)	19000	10	0.015	60	168
$mPEG_{5000}$-*b*-p((20%HEMAm-Lac4)-(80%HEMAm-Lac_2))	24000	6	0.08	80	8

** CMC = critical micelle concentration*

Encapsulation silicon phthalocyanine

A newly derivatized silicon phthalocyanine (SiPc) with maximum absorbance at 674 nm and high photocytotoxicity (IC_{50} = 0.1 µM [4]) was entrapped in $mPEG_{5000}$-***b***-p(HPMAm-Lac_2) micelles (Table 2).

Table 2: Effect of incorporation of a silicon phthalocyanine on the particle size of $mPEG_{5000}$-*b*-p(HPMAm-Lac_2) micelles and the encapsulation efficiency.

SiPc (mg/mL)	Z_{Ave} (nm)	PD	% SiPc encapsulated
-	64	0.01	-
0	78	0.07	-
0.5	74	0.24	84
1.5	*	*	69
2.5	*	*	61

** Could not be measured due to absorption of the DLS laser light by the SiPc.*

Whereas the solubility of the free phthalocyanine in water is negligible, these micelles were capable of solubilizing 0.15 mg phthalocyanine per mL.

pH dependent release of phthalocyanine

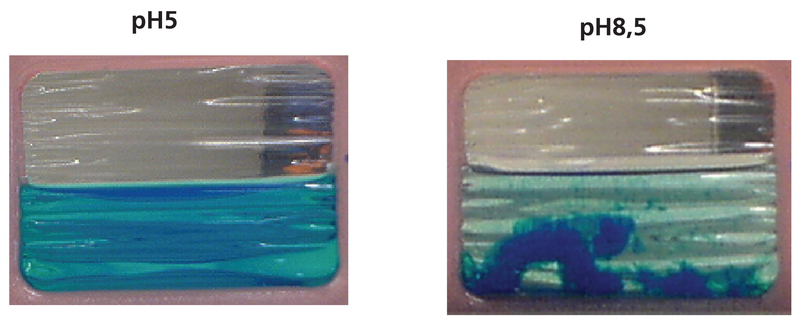


Figure 4 pH dependent release of a silicon phthalocyanine after 10 hours of incubation at 37°C.

In vitro studies show that at a low pH where the micelles are stable, the photosensitizer remained in the micellar core (fig 4, left). On the other hand at pH 8.5, where rapid hydrolysis of the lactic acid side chains occurs, the micelles destabilize which is associated with release of the phthalocyanine (fig 4 right).

Conclusion

- R1-PEG_{5000}-*b*-p(HE/PMAmLac$_n$) are thermosensitive amphiphilic block copolymers which form micelles (60-80 nm) above the cloud point.
- This carrier system was capable of encapsulating a hydrophobic silicon phthalocyanine up to a final concentration of 0.15 mg/mL.
- The phthalocyanine was only released once the micelles were destabilized.
- Further research involves biodistribution studies of the photosensitizers in the (targeted) micelles after i.v. administration and the therapeutic efficacy of the photosensitizer-loaded targeted micelles

References

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