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The relevance of hydrophobic segments in multiblock copolyesterurethanes for their enzymatic degradation at the air-water interface

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Abstract

The interplay of an enzyme with a multiblock copolymer PDLCL containing two segments of different hydrophilicity and degradability is explored in thin films at the air-water interface. The enzymatic degradation was studied in homogenous Langmuir monolayers, which are formed when containing more than 40 wt% oligo(*ε*-caprolactone) (OCL). Enzymatic degradation rates were significantly reduced with increasing content of hydrophobic oligo(*ω*-pentadecalactone) (OPDL). The apparent deceleration of the enzymatic process is caused by smaller portion of water-soluble degradation fragments formed from degradable OCL fragments. Beside the film degradation, a second competing process occurs after adding lipase from *Pseudomonas cepacia* into the subphase, namely the enrichment of the lipase molecules in the polymeric monolayer. The incorporation of the lipase into the Langmuir film is experimentally revealed by concurrent surface area enlargement and by Brewster angle microscopy (BAM). Aside from the ability to provide information about the degradation behavior of polymers, the Langmuir monolayer degradation (LMD) approach enables to investigate polymer-enzyme interactions for non-degradable polymers.

Keywords: Multiblock copolymer, Enzymatic polymer degradation, Oligo(ω-pentadecalactone), Oligo(ε-caprolactone), Langmuir monolayer degradation technique

1. Introduction

Polymeric biomaterials are accepted candidates for medical applications because of their easily tunable properties like mechanical and degradation characteristics. However, for clinical use new tailored materials are needed. Copolyesters such as poly(ω-pentadecalactone-*co*-*p*-dioxanone) consisting of hydrophobic and hydrophilic segments exhibit a high potential for medical applications since both biodegradation and physical properties can be tuned over a wide range by adjusting the polymer composition [1,2]. The relevance of poly(ω-pentadecalactone) (PPDL) as hydrophobic "PE-like" component is exhibited by several polymer systems such as PPDL-*b*-poly(*L*-lactic acid) (PLLA) copolymers or copolymers with different ε-substituted lactones [3-5]. Copolymers based on PPDL and δ-hexalactone are an alternative to poly(ε-caprolactone) (PCL) due to their lower stiffness and faster biodegradability [6]. Multiblock copolymers (PDLCL) consisting of two crystallizable segments, oligo(ε-caprolactone) (OCL) and oligo(ω-pentadecalactone) (OPDL) coupled by 2,2,(4),4-trimethylhexamethylene diisocyanate (TMDI) linkers are materials with good shape-memory capability [7,8]. By different temperature-memory catheter concepts the applicability of PDLCL multiblock copolymers for medical devices was investigated.

Thereby, the high potential for applications as minimal invasive devices was underlined due to the outstanding temperature-memory properties [7].

Among strategies to modify the degradation pattern of polymers, the synthesis of multiblock (co)polymers is promising [9,10]. Hereby, the degradation rate can be controlled by the weight content of, for example, crystallizable hard segments and hydrolyzable weak segments. PPDL is more resistant to hydrolytic degradation compared to other polyesters, like

PCL and PLLA, due to its higher melting temperature of the crystals and the lower amount of hydrolysable ester functions in the polymer backbone. However, the copolymerization of ω-pentadecalactone (PDL) and a variety of smaller lactones enables the synthesis of materials with controllable degradation properties (alkaline catalyzed) and a lower crystalline fraction [11,12]. OCL is also a favored material for the synthesis of (multiblock) copolymers, whereby the influence of the second block on the degradation behavior and on the mechanical and crystallization properties is confirmed in bulk studies [13-18]. For both homopolymers, PCL and PPDL, long degradation times are observed in hydrolytic bulk degradation studies (pH = 7.4) resulting from high crystallinity and hydrophobic character [19,20].

In 3D polymer samples the diffusion rate of water strongly affects the degradation kinetics especially in the case of highly hydrophobic materials. Therefore, methods are needed which can separate these transport-phenomena from the involved chain scission process. The Langmuir monolayer degradation (LMD) technique enables such separation and allows insights in the degradation mechanism on the molecular level [21-24]. In LMD studies the decrease of the surface areas of spread polymeric monolayers are recorded under isobaric conditions and directly correlated to degradation processes. During the degradation of the polymeric films short water-soluble fragments are formed. The degradation of PCL is accelerated in bulk studies as well as in 2D monolayer investigations by different types of lipases, for instance the lipase from *Porcine* pancreas [25], *Pseudomonas alcaligenes* [26], or *Pseudomonas cepacia* [27]. Lipases are able to hydrolyze triacylglycerols and other ester bonds. Their interfacial activity results in conformational alteration, which leads to an exposition of its hydrophobic surface to the substrate [28]. In multiblock copolymers composed of poly(*p*-dioxanone) (PPDO) and PCL segments (PDC) linked by TMDI, the 2D enzymatic degradation behavior of PCL can be tuned by using different compositions [29]. For those multiblock copolymers surface erosion mechanisms with linear enzymatic bulk degradation kinetics is verified in 3D experiments [30]. At the air-water interface the

enzymatic degradation kinetics was described by the random chain scission mechanism. In this case, the formation of water-soluble degradation fragments of the PPDO segments accelerates the fast diffusion of the degradation fragments out of the material. In contrast, the hydrophobic OPDL segments in PDLCL are water-insoluble, which makes LMD experiments for degradation studies questionable. However, PDLCL multiblock copolymers form homogeneous Langmuir films and thus fulfill the necessary requirement of an extended chain conformation, which makes LMD experiments feasible. We hypothesize that the enzymatic degradation behavior of PDLCL multiblock copolymers containing hydrophobic OPDL blocks is strongly influenced by the polymer composition. PPDL cannot be degraded by the lipase from *Pseudomonas cepacia* in bulk studies [19] and does not form water-soluble degradation fragments in the investigation time period. Therefore, it is expected that the variation of OPDL content influences the enzymatic degradation kinetics of these polymers at the air-water interface.

In this manuscript, the influence of a hydrophobic OPDL segment on the degradation of three PDLCLs differing in their composition is investigated and compared to the degradation behavior of OCL at the air-water interface. Langmuir layers from multiblock copolymers containing degradable OCL segments are expected to result in a faster surface area reduction. The Langmuir layer properties of low molecular weight OPDL are studied in comparison complementary under isobaric conditions including the interaction with the lipase from *Pseudomonas cepacia* whereby the morphological behavior is visualized by BAM. LMD experiments are also expected to elucidate the hydrophobic polymer-enzyme interactions and contribute to a better insight into degradation mechanisms.

2. Experimental Part

2.1 Materials

The PDLCL multiblock copolymers were synthesized via co-condensation of oligo(*ω*-pentadecalactone) macrodiols (OPDL, M_n = 2400 g·mol^{-1}), which was prepared by ring-opening polymerization of *ω*-pentadecalactone (PDL, Macrolid Supra, Th. Geyer, Friedrichsthal, Germany) with 1,8-octanediol as initiator, and oligo(*ε*-caprolactone) macrodiols (OCL, M_n = 2400 g·mol^{-1}, trade name CAPA2304, Solvay Caprolactones, Warrington, U.K.) with 2,2,(4),4-trimethylhexamethylene diisocyanate (TMDI, Sigma-Aldrich, Germany) as described elsewhere [7] (Scheme 1).

The OCL-content in the starting material composition, the number-average molecular weights (M_n), the dispersity index (PDI), and the mean molecular weight of the repeating unit (MMW_{RU}: calculated accordingly as sum of the molecular weights of *ε*-caprolactone, *ω*-pentadecalactone, and TMDI each multiplied with the respective weight percentage number) for determining the mean molecular area of the repeating unit (MMA) are listed in Table 1. All polymers were used without any further purification and chloroform (HPLC grade purity, Roth, Germany) was applied as spreading solvent. As subphase for Langmuir monolayer experiments deionized water (18.2 MΩ·cm) purified by a Milli-Q Gradient A-10 water purification system (Millipore, Merck, Germany) was used.

Table 1 Number-average molecular weights M_n, the dispersity index (DI), the mean molecular weight of the repeating unit (MMW_{RU}) and composition of the PDLCLs, OCL and OPDL.

Sample ID [a]	**OCL-content [mol%]**	**MMW_{RU} [g mol^{-1}]**	**M_n** [b] **[g mol^{-1}]**	**DI** [b]
OPDL	0	240	2700	1.7
PDLCL40	58	178	86,000	3.3
PDLCL50	68	159	59,000	2.0
PDLCL70	83	137	116,000	2.7
OCL	100	114	2800	1.3

[a] The two-digit number gives the weight content of OCL in wt% of the starting composition.
[b] For PDLCL the M_n and DI were abstracted from literature measured by GPC [31] and for OCL and OPDL the values were determined by GPC (universal calibration) using 250 mm x 4.6 mm GRAM gel columns, 3 and 3 x 10^2 nm porosity, 10 µm particle size (Polymer Standard-Service GmbH, Mainz, Germany, PSS), a degasser (ERC-3315, Riemerling, Germany), a gradient pump PU 980 and an automatic injector AS-851 (both Jasco, Tokyo, Japan) and $CHCl_3$ as eluent at 35°C with a flow rate of 0.25 mL·min^{-1}.

2.2 Methods

Two different KSV Nima troughs (KSV Nima, Finnland) were used, a "High compression" for recording the surface pressure-area isotherm and Langmuir monolayer degradation studies and a "Large" trough for recording Brewster angle microscopy (BAM) images, both provided with a Wilhelmy type dynamometric system using a strip of filter paper located equidistant between two movable barriers. BAM images of the film morphology were obtained by BAM of maximum 500 × 400 µm² in size using an ellipsometer nanofilm_ep3 (EP3, Accurion, Göttingen, Germany) with a wavelength of 658 nm, which can record with a lateral resolution of 2 µm. BAM micrographs were taken with a ×10 magnification lens and a high performance CCD camera (765 x 572 pixel). The monolayers were compressed by a constant compression rate of 10 mm min^{-1}. The microscope and the Langmuir film micro-balance were located on a table with vibration isolation (halcyonics variobasic 40, Accurion, Göttingen, Germany). The surface pressure is recorded as a function of the mean molecular area per repeating unit (MMA). All presented isotherm data were reproducible with a random measurement error of

5% concerning the surface pressure or the MMA values for three independently repeated experiments. The standard deviation in the degradation curves for PDLCL is about ± 0.1 a. u..

The trough and the barriers were cleaned thoroughly with chloroform and filled with deionized water. The cleanliness of the trough and the subphase was controlled by monitoring the surface pressure, while closing the barriers. The total change of the surface pressure π should be below $\Delta\pi \leq 0.2$ mN·m^{-1}. For all samples the spreading solutions had a concentration between ~ 0.26 and 0.54 mg·mL^{-1}. A microsyringe (Hamilton Co., Reno, NV, USA) was used to apply the spreading solution drop-wise onto the air-water interface.

Langmuir monolayer degradation (LMD) experiments were performed at 22 °C and 37 (± 0.5) °C. For high subphase temperature experiments the temperature was kept constant by a thermostat and was adjusted for at least 1 h. LMD experiments were performed using a handcrafted level compensation setup to overcome the problem of intensive subphase evaporation influencing the film micro-balance performance. For enzymatic degradation experiments, a phosphate buffered (PBS, pH = 7.4, Omnilab, Germany) subphase was used. After spreading the polymer solution and evaporation of the $CHCl_3$ for 10 min, the polymer film was compressed under constant rate compression conditions to a defined target surface pressure (π = 7.0 mN·m^{-1}), which was kept constant for at least 30 min before the lipase solution was injected (V = 4.5 mL, c = 1 mg·mL^{-1}, V_{trough} ~ 450 mL). The surface pressure was kept constant by movable barriers. A measuring unit for the release of low molecular fragments into the subphase is the corrected surface area reduction $\Delta A_{corr}(t)$ [22]. It was calculated according to equation (1), where A_t is the reduced mean molecular area per repeating unit at the time t and A_0 (A(t = 0) is the surface area initially occupied by the compressed Langmuir film:

$$\Delta A_{corr}(t) = (A_0 A_t^{-1})\text{-}1 \quad (1)$$

3. Results and Discussion

3.1 Langmuir monolayer isotherm experiments

The urethane free OPDL cannot be spread at the air-water interface and does not form Langmuir monolayers. Its surface behavior is consistent with that of previously investigated polyesterurethanes OPDL-TMDI [32]. BAM investigation confirms that the increase in the surface pressure is caused by the compression of single slabs, which form a rather stiff layer on the water surface (not shown). The observed precipitate-like structures of OPDL clearly exhibit the more hydrophobic character of OPDL compared to OCL, which is spreadable on the water surface.

On the contrary, PDLCLs with a content of above 40 wt% OCL form highly compressible stable films at the air-water interface [32]. The surface pressure-area (π-*A*) isotherms indicate transition regions which are typical for the coexistence of liquid expanded (LE) and liquid condensed (LC) phases and becomes broader with increasing OCL content (PDLCL50: 7.5 - 20 Å²; PDLCL70: 5 -22 Å²) (Fig. 1) [33,34]. Homogeneous films are revealed by BAM images in the semi-diluted state for all investigated polymers allowing to perform LMD studies.

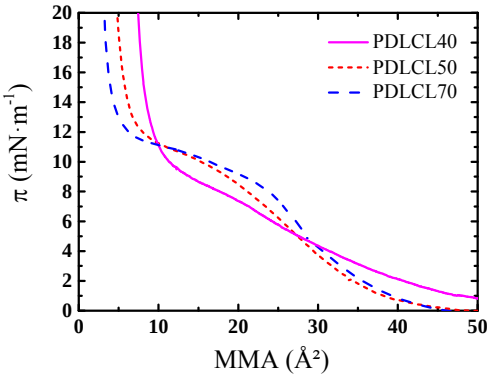


Fig. 1 π-*A* isotherm of PDLCL Langmuir films (drawn through line: PDLCL40; short dashed line: PDLCL50; dashed line: PDLCL70) on a pure water subphase at 22 °C

3.2 Langmuir Monolayer Degradation Experiments

For the enzymatic LMD experiments the lipase from *Pseudomonas cepacia* was chosen which is known for its ability to degrade PCL and for its inability to degrade PPDL in bulk studies [19]. The LMD investigations at a surface pressure 7 $mN \cdot m^{-1}$ are performed at 37 °C on PBS buffered subphase (pH = 7.4). Fig. 2 shows the corrected surface area reduction against the reaction time, the so-called degradation curves.

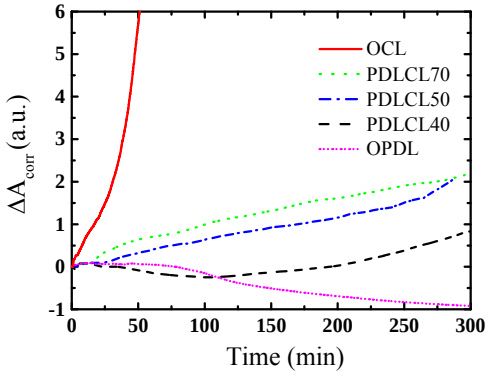


Fig. 2 Correlated surface area reduction (ΔA_{corr}) against the time determined in a Langmuir monolayer degradation experiment under isobaric conditions ($\pi = 7$ $mN \cdot m^{-1}$) of OCL, PDLCL70, PDLCL50, PDLCL40, and OPDL with the lipase from *Pseudomonas cepacia* at 37 °C on a PBS buffered subphase. The time t = 0 min represents the injection of the lipase into the subphase

The degradation rates for PDLCL-based Langmuir films increase with increasing OCL content, but, even PDLCL70 shows a significant reduced formation of water-soluble degradation fragments compared to the OCL oligomer. The considerably reduced degradation rates can be caused by the molecular weights, by differences in the hydrolysis rates in the segments, or by the presence of the urethane junction units. It is known that low molecular OCL degrades faster than high molecular PCL, but PCL with M_n 80.000 $g \cdot mol^{-1}$ is fully degraded after approximately 1 h under comparable experimental conditions [22]. Therefore, effects of the molecular weight can be excluded for the observed retarded degradation. Distinctions in the hydrolysis of the different polyester backbones can also be neglected for

several reasons. Both PCL and PPDL are slowly hydrolytically degradable in bulk [19,35]. At the air-water interface the hydrolytic degradation of both homopolymers and PDLCL films is also found to be very slow [22,32]. For example, the formation of water-soluble degradation fragments by hydrolysis of PCL (M_n = 10.000 g·mol^{-1}) occurs with $\Delta A_{corr} \sim 0.1$ per hour [22]. Since all investigated PDLCL samples exhibit the same ratio of polymer segments to linker units, the influence of the urethane junction unit on the degradation rate can also be neglected. Considering these facts, the degradation of the PDLCL films can only correlate with the polymer composition. A necessary prerequisite for the application of the LMD approach is the ability of polymers to form water-soluble degradation fragments in the experimental time period. Since only water-soluble OCL fragments are formed, a defined surface area remains after the cleavage of all degradable OCL bonds. The limiting values (ΔA_{corr}(lim)) are listed in Table 2.

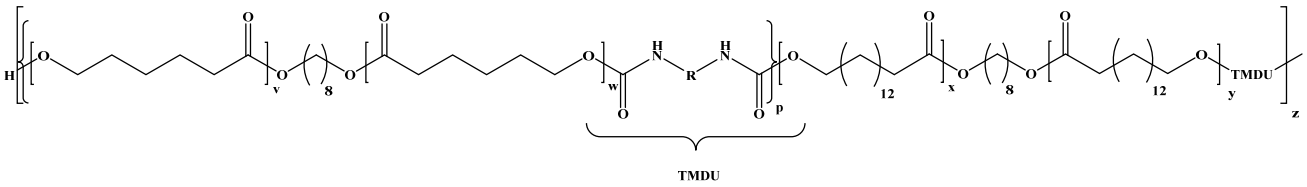
Table 2 Limiting values of formed water-soluble fragments (ΔA_{corr}(lim)) for PDLCL and OPDL calculated on the basis of the remaining area after the complete degradation of all OCL segments and their dissolving into the subphase and the experimental determined ΔA_{corr} values after a time interval of 5 h (PDLCL50: 4 h)

Sample ID	**Remaining Area relating to wt% [%]**	**ΔA_{corr}(lim)**	**ΔA_{corr} after 5 h**
OPDL	100	0[a]	-1.00 ± 0.3[b]
PDLCL40	60	0.66	0.9 ±0.3
PDLCL50	50	1.00	1.2 ± 0.1
PDLCL70	30	2.33	1.8 ± 0.3

[a]OPDL cannot be degraded hydrolytically or enzymatically under the experimental conditions.
[b]Negative ΔA_{corr}(lim) values refer to an increase of the required surface are, e. g. by incorporation of enzymes.

The variance between the experimental and limiting values might be explained by the dispersity and segment distribution of the polymer samples. This approach shows that the

lower OCL content in PDLCL leads to a reduced formation of water-soluble degradation fragments (reduced ΔA_{corr} value), which has to be considered in further mechanistical interpretations of the degradation process. But, the reduced limiting ΔA_{corr} values do not explain the significantly longer time periods observed for the degradation of PDLCL films.



Scheme 1 Chemical structure of multiblock copolyesterurethanes based on oligo(*ω*-pentadecalactone) and oligo(*ε*-caprolactone) (PDLCL).

As mentioned above, the degradation of PDLCL films by *Pseudomonas cepacia* lipase occurs in the OCL segments, only. Solely, the produced small, water-soluble OCL fragments (tetramer or smaller) [24], which are not connected to hydrophobic OPDL segments, can leave the interface to the subphase thereby contributing to a surface area reduction (Scheme 2a). But, the solubility of the OCL degradation fragments is modified by the significant hydrophobicity of OPDL blocks. The probability to generate water-soluble degradation fragments decreases with increasing amount of hydrophobic OPDL segments [24]. Each OCL segment or monomer unit, which is connected to an OPDL block stays at the air-water interface until this specific linking ester bond is cleaved. For long experimental periods the formation of water-insoluble OPDL degradation fragments cannot be excluded but, even the smallest OPDL degradation products (15-hydroxypentadecanoic acid and its oligomers) stay at the air-water interface because of their water-insolubility. So, even the less probable hydrolysis in OPDL blocks does not contribute to the surface area reduction.

The PDLCL multiblock copolymer is composed of degradable OCL and hydrophobic segments, which are water-insoluble and non-degradable by the lipase from *Pseudomonas cepacia*. The question arises, if the degradation curve would be changed in case of non-degradable but water-soluble segments. Reiche et al. showed for PDC multiblock copolymers the influence of a water-soluble block segment on degradation whose degradation kinetics is almost a linear function of time [29]. In this case, the PPDO blocks are anchored at the air-water interface by the more hydrophobic PCL blocks. But, due to the high hydrophilicity of PPDO, the cleavage of only two (in an extreme case only one) PCL ester bonds is sufficient to generate large, water-soluble degradation products based on PPDO with "long" PCL chains. In Scheme 2b the fast formation of water-soluble degradation fragments is shown, which can explain the experimentally observed linear surface area reduction. The comparison of PDLCL and PDC multiblock copolymer systems containing enzymatically non-degradable segments (ODPL or PPDO), show that the evaluation of the experimental monolayer degradation data cannot be revealed by the mathematical dynamic fragmentation model (Fig. 3).

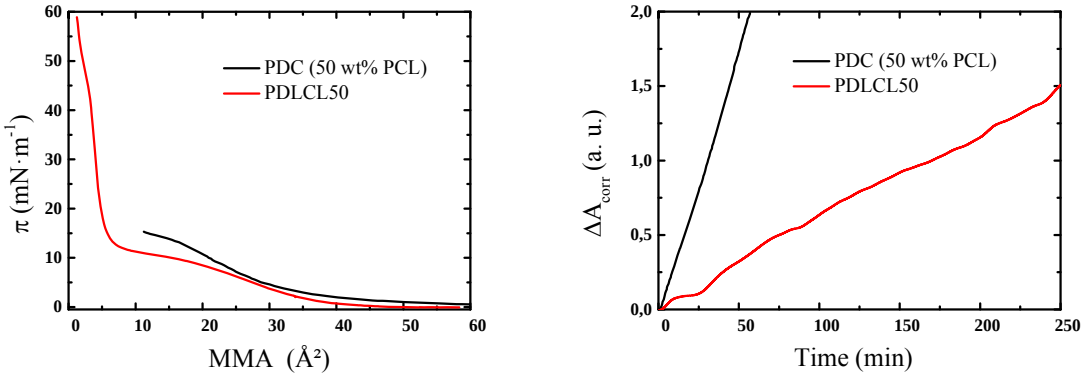
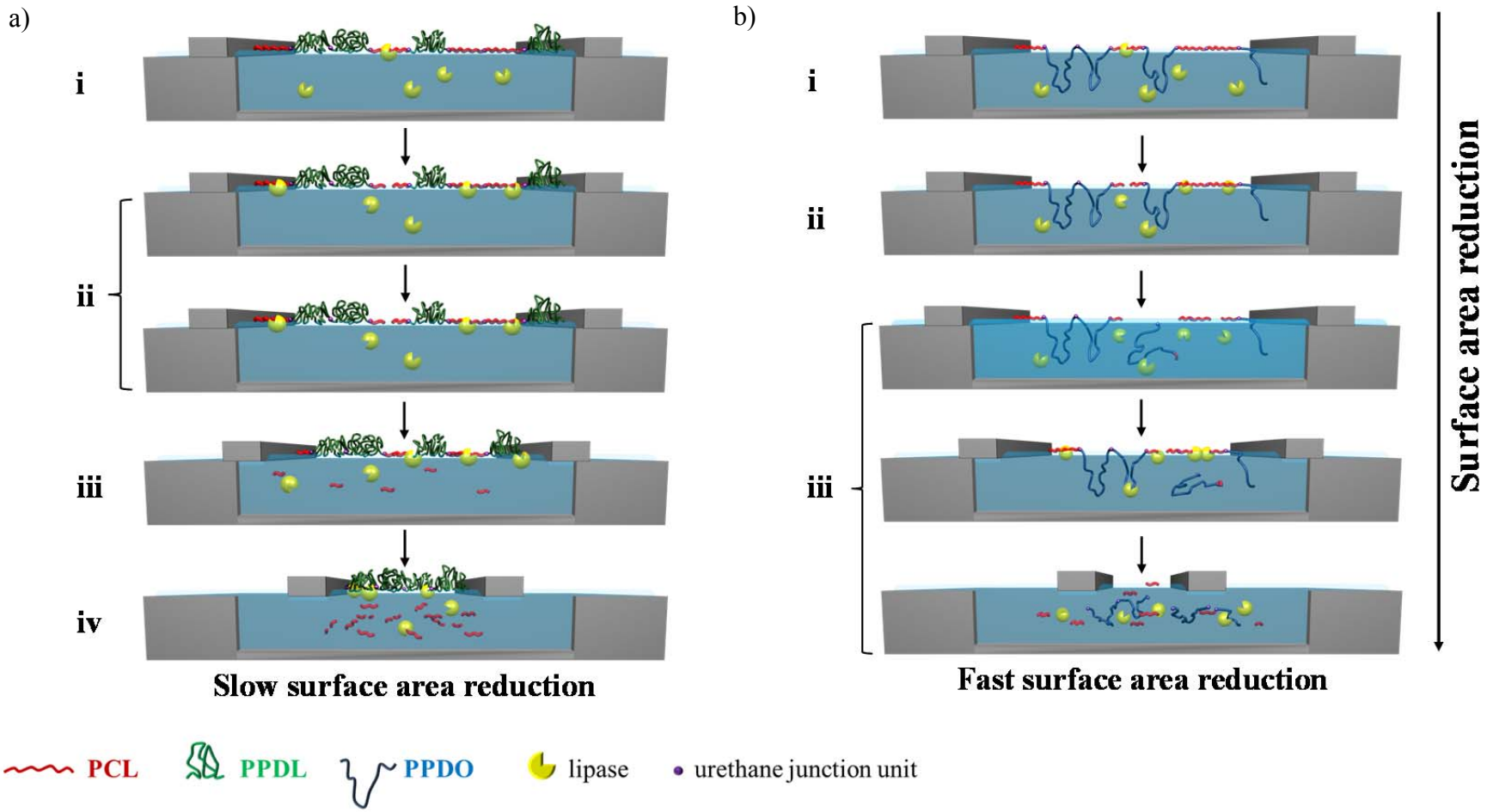


Fig. 3 a) π-*A* isotherm and b) degradation curve of PDLCL50 and PDC (50 wt% PCL). Data for the PDC curves were extracted from Ref. [29].

Scheme 2 Illustration of the surface area reduction of OCL-based multiblock copolymers using a) hydrophobic and b) hydrophilic segments. Schema 2a: i) the injection of the enzyme and polymer-enzyme interaction, ii) the fragmentation into water-insoluble products, iii) the formation of small water-soluble products, and iv) the formation of a hydrophobic layer. Schema 2b: i) the injection of the enzyme and polymer-enzyme interaction, ii) the formation of more hydrophilic segments, and iii) the formation of large water-soluble products



3.3 Polymer-Enzyme Interaction

The processes shown in Scheme 2 do not completely explain the low ΔA_{corr} values and the delay time period in the degradation curves of the PDLCL films. To elucidate the degradation behavior of the Langmuir films by LMD the behavior of PDLCL40 and PDLCL50 including the highest amounts of OPDL segments are investigated in more detail. The surface area for PDLCL40 slightly increases (negative ΔA_{corr} values) before the degradation of the polymer films starts. For OPDL almost constant ΔA_{corr} values are obtained in the first period of the experiments (~1 h) and an ongoing increase of the surface area occurs in the second period (Fig. 2). The slope of the surface area-time curve for OPDL clearly illustrates the existence of a second process apart from OCL degradation during the LMD experiments for PDLCL.

To obtain *in situ* information of the polymer-enzyme interaction and its role in reflecting the degradation kinetics at the air-water interface, time-dependent BAM measurements are performed simultaneously to the Langmuir monolayer (degradation) experiments (π = 7 mN·m ^{1}) using OPDL and PDLCL50. The trough area changes are shown in Fig. 4b for OPDL. After injection of the lipase solution into the subphase, the homogeneous film (Fig. 4a) starts to rupture at several positions (Fig. 4c, image i) and small slabs are formed. With ongoing time, the slabs become smaller and the unstructured interface appears in the (almost black) background (Fig. 4c, image ii). The surface area increases slowly reaching equilibrium after 6 h with ongoing morphological structure formation (Fig. 4c, images iii and iv). Surprisingly, these significant morphological changes occur although during the first hour almost no change in the surface area is observed. Considering the film thickness (~15 nm) of OPDL-TMDI at the air-water interface [32], probably a displacement of water by the enzyme occurs resulting in film extension after a longer period of time (> 1 h). Constituting the size of the lipase by using crystal structure data the amount of lipase in the subphase solution (~10^{17} molecules) is higher than the required amount of lipase (~10^{15} molecules) necessary for

the observed increase in surface area of about 300 cm² [36]. That means that the surface area enlargement can be associated with the polymer-enzyme interaction at the interface although BAM images revealed a reduced surface area coverage.

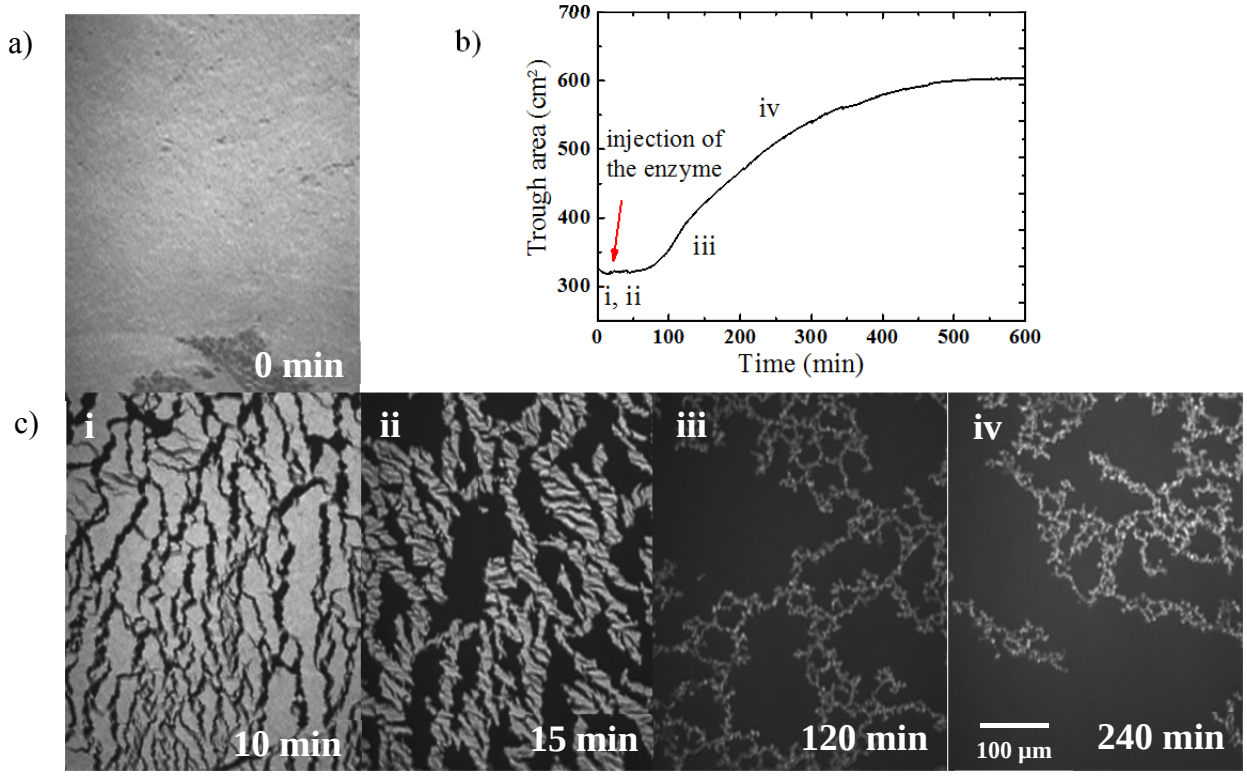


Fig. 4 Langmuir monolayer experiment under isobaric conditions ($\pi = 7$ mN·m^{-1}) of OPDL on a PBS buffered subphase at 22 °C by adding *Pseudomonas cepacia* lipase (marked with the red arrow). a) BAM image before the injection of the lipase, b) graph of the trough area against the time, and c) BAM images at different time points after lipase injection

Small holes appear in the initially homogeneous PDLCL50 film after one hour, which could indicate OCL degradation. However, pronounced morphological changes are not observed after a degradation time of 3 h as observed for OPDL films. Consequently, besides degradation the surface-active lipase is able to be incorporated into the polymer layer thereby effecting the interface morphology. This elucidates the difficulties using the LMD approach for copolymers with a hydrophobic, non-degradable polymer block.

Temperature-dependent degradation experiments are performed to get an insight into the two competitive processes: polymer chain scission and enrichment of the enzyme at the interface.

In Fig. 5 the degradation curves for PDLCL50 at 22 °C and 37 °C are shown. A slower enzymatic hydrolysis at 22 °C compared to 37 °C is observed as expected from the investigation of Kulkarni et al., who measured an increased enzyme activity of the lipase from *Pseudomonas cepacia* with increasing temperature [22]. For experiments at 22 °C the enrichment of the lipase in the polymer-water interface is the predominant process at the beginning of the experiment. Only after a longer delay time period a sufficient amount of ester bonds are cleaved to generate low molecular fragments, which can leave the interface followed by a surface area reduction. At 37 °C the hydrolysis proceeds faster immediately after injection despite a lower amount of lipase at the interface. The comparison of the two degradation curves at different temperatures reflects the two ongoing processes. On the one hand, the surface activity of the lipase, which leads to an apparent stagnation in the formation of water-soluble degradation fragments at 22 °C, and on the other hand, the surface area reduction due to polymer degradation, which is predominant at 37 °C already in the first stage.

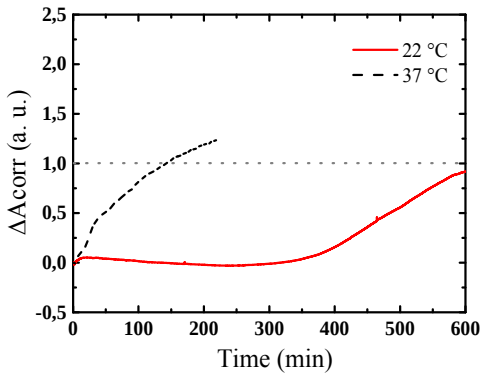


Fig.5 Langmuir monolayer degradation of PDLCL50 with the lipase from *Pseudomonas cepacia* at 22 °C and 37 °C determined under isobaric conditions (π = const. = 7 mN·m^{-1}) figured as correlated surface area reduction (ΔA_{corr}) against the time (the dotted line represents the limiting value of formed water-soluble OCL fragments according to Table 2)

4. Conclusion

The enzymatic degradation behavior is investigated for Langmuir monolayers based on multiblock copolymers containing different degradable segments namely OCL and hydrophobic OPDL segments with varying compositions. The presence of hydrophobic OPDL blocks significantly reduces the enzymatic degradation rate of PDLCL film by lipase from *Pseudomonas cepacia*. The OCL segment degradation is retarded due to the circumstance that only small OCL degradation fragments (up to tetramers), which are not connected to an OPDL segment can leave the interface.

Two competing processes are identified by LMD experiments after adding the lipase into the subphase, namely the polymer degradation by the formation of water-soluble fragments and the enrichment of the lipase molecules into the polymeric monolayer. Both effects can be separately verified in temperature-dependent measurements. The enrichment of the enzyme in the polymer-water interface is confirmed in independent experiments using low-molecular weight OPDL by film surface area enlargement and visualized by BAM. With increasing OPDL content the interaction between PDLCL and enzyme increases.

The presented results show for the first time that for multiblock copolymers only from the slope of the degradation curve, the molecular degradation mechanism as chain-end cut or random chain scission cannot be deducted. Beside the degradation experiments, the approach of the Langmuir experiments enables the investigation of the polymer-enzyme interactions for non-degradable polymers. For OPDL significant morphological changes are observed due to the enrichment and incorporation of the lipase into the polymer layer.

Based on the knowledge generated from our LMD experiments we expect that also in time-consuming enzymatic bulk studies the degradation by lipase from *Pseudomonas cepacia* is retarded for PDLCL in comparison to PCL. Moreover, a modified erosion mechanism can be

expected due to the ability of the lipase to penetrate into the hydrophobic OPDL parts of the PDLCL matrix.

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