University of Groningen

Dipping-Induced Azimuthal Helix Orientation in Langmuir-Blodgett Monolayers of alpha-Helical Amphiphilic Diblock Copolypeptides

Nguyen, Le-Thu T.; Vorenkamp, Eltjo J.; ten Brinke, Gerrit; Schouten, Arend J.

Published in:
Langmuir

DOI:
[10.1021/la1002622](https://doi.org/10.1021/la1002622)

IMPORTANT NOTE: You are advised to consult the publisher's version (publisher's PDF) if you wish to cite from it. Please check the document version below.

Document Version
Publisher's PDF, also known as Version of record

Publication date:
2010

[Link to publication in University of Groningen/UMCG research database](#)

Citation for published version (APA):

Nguyen, L.-T. T., Vorenkamp, E. J., ten Brinke, G., & Schouten, A. J. (2010). Dipping-Induced Azimuthal Helix Orientation in Langmuir-Blodgett Monolayers of alpha-Helical Amphiphilic Diblock Copolypeptides. *Langmuir*, 26(13), 11018-11024. <https://doi.org/10.1021/la1002622>

Copyright

Other than for strictly personal use, it is not permitted to download or to forward/distribute the text or part of it without the consent of the author(s) and/or copyright holder(s), unless the work is under an open content license (like Creative Commons).

The publication may also be distributed here under the terms of Article 25fa of the Dutch Copyright Act, indicated by the "Taverne" license. More information can be found on the University of Groningen website: <https://www.rug.nl/library/open-access/self-archiving-pure/taverne-amendment>.

Take-down policy

If you believe that this document breaches copyright please contact us providing details, and we will remove access to the work immediately and investigate your claim.

Downloaded from the University of Groningen/UMCG research database (Pure): <http://www.rug.nl/research/portal>. For technical reasons the number of authors shown on this cover page is limited to 10 maximum.

Dipping-Induced Azimuthal Helix Orientation in Langmuir–Blodgett Monolayers of α -Helical Amphiphilic Diblock Copolypeptides

Le-Thu T. Nguyen, Eltjo J. Vorenkamp, Gerrit ten Brinke, and Arend J. Schouten*

Department of Polymer Chemistry, Zernike Institute for Advanced Materials, University of Groningen, Nijenborgh 4, 9747 AG Groningen, The Netherlands

Received January 19, 2010. Revised Manuscript Received April 16, 2010

The azimuthal helix orientation of the rigid-rod amphiphilic diblock copolypeptides (PLGA-*b*-PMLGSLGs) of poly(α -L-glutamic acid) (PLGA) and poly(γ -methyl-L-glutamate-*ran*- γ -stearyl-L-glutamate) with 30 mol % of stearyl substituents (PMLGSLG) in Langmuir–Blodgett (LB) monolayers was investigated using polarized transmission Fourier transform infrared spectroscopy. The relative position of dipping with respect to the previous transfer position can be used to manipulate the azimuthal orientation of the helices parallel to or tilted by an angle of 45° with respect to the dipping direction in the transferred films. The study of the azimuthal order for the LB monolayers of PLGA-*b*-PMLGSLGs of various block lengths revealed that the observed effect arises mainly from the deformation of the PMLGSLG top brush layer, induced by the flow orientation around the transfer region. In those cases where the PMLGSLG block is tilted by a sufficiently large angle with respect to the surface normal, high azimuthal order parameters of 0.5–0.75 were obtained.

Introduction

The Langmuir–Blodgett (LB) technique has proven to be a powerful tool for the creation of ultrathin films with control over two-dimensional order and packing of molecules.¹ Especially, in-plane preferential molecular alignment can be introduced in the LB films due to the orientation phenomenon during the vertical transfer of Langmuir films onto solid substrates. This special orientation effect has been observed for rigid-rod polymers,^{2,3} rod-like crystals,⁴ stacks of disk-like molecules^{5,6} and stretched two-dimensional flexible polymer networks.⁷ The in-plane anisotropy in LB films is of great interest as it offers the materials useful physical properties for potential applications as alignment layers of liquid crystals,^{8,9} optical nonlinear waveguides,^{10,11}

matrices for orienting conductive guest molecules,¹² and electro-^{13–21} and microelectronic materials.

The reported orientation phenomenon during the transfer process results in an alignment of the molecular main-chain axis along the dipping direction. The effect has been explained by flow orientation models,^{22–24} in which a sink flow near the transfer region is responsible for orientation of the molecules parallel to the transfer direction. It has been reported for hydrophobic rigid-rod polymers and aggregates that the transfer speed and substrate width with respect to the trough width are main factors that affect the degree of orientation.^{6,22,23}

Hairy-rod polypeptides provide stable LB films with a highly ordered array of α -helices aligned parallel to the substrate.² They show lyotropic and thermotropic behavior^{25,26} and have received attention in the past years.^{27,28} LB films of these hairy-rod polymers show strong dipping-induced in-plane anisotropy with a very high degree of orientation parallel to the dipping direction, which has been demonstrated both experimentally and theoretically.^{22,29}

We studied amphiphilic diblock copolypeptides consisting of a hydrophobic block of hairy-rod poly(γ -methyl-L-glutamate-*ran*- γ -stearyl-L-glutamate) with 30 mol % of stearyl substituents

*Corresponding author. E-mail address: A.J.Schouten@rug.nl.

(1) Zasadzinski, J. A.; Viswanathan, R.; Madsen, L.; Garnæs, J.; Schwaetz, D. K. *Science* **1994**, *263*, 1726.
(2) Duda, G.; Schouten, A. J.; Arndt, T.; Lieser, G.; Schmidt, G. F.; Bubeck, C.; Wegner, G. *Thin Solid Films* **1988**, *159*, 221.
(3) Sauer, T.; Arndt, T.; Batchelder, D.; Kalachev, A. A.; Wegner, G. *Thin Solid Films* **1990**, *187*, 357.
(4) Brinkhuis, R. H. G.; Schouten, A. J. *Macromolecules* **1992**, *25*, 2717.
(5) Vandevyver, M.; Albouy, P.; Mingotaud, C.; Perez, J.; Barraud, A.; Karthaus, O.; Ringsdorf, H. *Langmuir* **1993**, *9*, 1561.
(6) Ogawa, K.; Yonehara, H.; Shoji, T.; Kinoshita, S.; Maekawa, E.; Nakahara, H.; Fukuda, K. *Thin Solid Films* **1989**, *178*, 439.
(7) Hagting, J. G.; Vorenkamp, E. J.; Schouten, A. J. *Thin Solid Films* **1998**, *327–329*, 65.
(8) Michinobu, T.; Nakada, K.; Matsuda, M.; Shigehara, K. *Colloids Surf., A* **2008**, *321*, 82.
(9) Lu, R. B.; Xu, K. S.; Zhang, Z. K.; Zhong, J. H.; Li, G. Z.; Wu, H. M.; Lu, Z. H. *Mol. Cryst. Liq. Cryst.* **1996**, *289*, 77.
(10) Bosshard, C.; Floersheimer, M.; Kuepfer, M.; Guenter, P. *Opt. Commun.* **1991**, *85*, 247.
(11) Han, K.; Wu, Y.; Tang, G.; Zhang, G.; Wang, Q.; Li, H. *Opt. Mater.* **2004**, *27*, 155.
(12) Vandevyver, M.; Bourgoin, J. P.; Barraud, A.; Perez, X.; Veber, M.; Jallabert, C.; Strezelecka, H. J. *Phys. D: Appl. Phys.* **1992**, *25*, 284.
(13) Mattu, J. S.; Leach, G. W. *J. Am. Chem. Soc.* **2010**, *132*, 3204.
(14) Okahata, Y.; Kobayashi, T.; Tanaka, K. *Langmuir* **1996**, *12*, 1326.
(15) Li, J.; Zhang, Y. *Carbon* **2007**, *45*, 493.
(16) García-Vázquez, P.; Morales-Saavedra, O. G.; Pelzl, G.; Guadalupe Bañuelos, J.; Carreón-Castro, M. P. *Thin Solid Films* **2009**, *517*, 1770.
(17) Mattu, J.; Johansson, T.; Holdcroft, S.; Leach, G. W. *J. Phys. Chem. B* **2006**, *110*, 15328.

(18) Piris, J.; Debije, M. G.; Stutzmann, N.; Laursen, B. W.; Pisula, W.; Watson, M. D.; Bjornholm, T.; Muellen, K.; Warman, J. M. *Adv. Funct. Mater.* **2004**, *14*, 1053.
(19) Laursen, B. W.; Norgaard, K.; Reitzel, N.; Simonsen, J. B.; Nielsen, C. B.; Als-Nielsen, J.; Bjornholm, T.; Solling, T. I.; Nielsen, M. M.; Bunk, O.; Kjaer, K.; Tchegbotareva, N.; Watson, M. D.; Muellen, K.; Piris, J. *Langmuir* **2004**, *20*, 4139.
(20) Tanaka, H.; Mizutani, T.; Kuroda, S. *Colloids Surf., A* **2006**, *284–285*, 97.
(21) Okahata, Y.; Kawasaki, T. *Top. Curr. Chem.* **2005**, *260*, 57.
(22) Schwiégk, S.; Vahlenkamp, T.; Xu, Y.; Wegner, G. *Macromolecules* **1992**, *25*, 2513.
(23) Minari, N.; Ikegami, K.; Kuroda, S.; Saito, K.; Saito, M.; Sugi, M. *Solid State Commun.* **1988**, *65*, 1259.
(24) Tabe, Y.; Ikegami, K.; Sugi, M. *J. Appl. Phys.* **1993**, *73*, 905.
(25) Watanabe, J.; Sakajiri, K.; Okoshi, K.; Kawachi, S.; Magoshi, J. *Macromol. Chem. Phys.* **2001**, *202*, 1004.
(26) Watanabe, J.; Fukuda, Y.; Gehani, R.; Uematsu, I. *Macromolecules* **1984**, *17*, 1004.
(27) Ando, I.; Yin, Y.; Zhao, C.; Kanesaka, S.; Kuroki, S. *Macromol. Symp.* **2005**, *220*, 61.
(28) Fukuto, M.; Heilmann, R. K.; Pershan, P. S.; Yu, S. M.; Soto, C. M.; Tirrell, D. A. *J. Chem. Phys.* **2003**, *119*, 6253.
(29) Duda, G.; Wegner, G. *Makromol. Chem., Rapid Commun.* **1988**, *9*, 495.

Scheme 1. Chemical Structure of PLGA-*b*-PMLGSLG and Simple Schematic Representation of the Double-Brush Structure of the PLGA-*b*-PMLGSLG LB Monolayer

(PMLGSLG) and a hydrophilic block of rigid-rod poly(α -L-glutamic acid) (PLGA). Besides the high mobility provided by the amorphous side chain mantle of the hairy-rod polypeptide block, monolayers of these diblock copolypeptides exhibit a brush structure composed of densely packed parallel α -helices, which are tilted at an angle with respect to the water surface normal.^{30–32}

Interestingly, we observed strong effects of in-plane anisotropy produced by the transfer process in the LB monolayers of these diblock copolypeptides. Particularly, a dipping-induced helix orientation oblique by an angle with respect to the dipping direction was observed for the first time. Here, the azimuthal helix orientation effect induced by the vertical transfer of LB monolayers of the PLGA-*b*-PMLGSLG diblock copolymers is reported.

Experimental Section

Materials. The PLGA-*b*-PMLGSLG diblock copolymers are abbreviated as CoPo-*m*-*n*, where *m* and *n* denote the degrees of polymerization of the PLGA and PMLGSLG blocks, respectively (Scheme 1). A detailed description of the synthesis of PLGA-*b*-PMLGSLGs can be found elsewhere.³⁰ In short, α -helical PLGA-*b*-PMLGSLG was synthesized via a diblock copolymer precursor consisting of poly(γ -*tert*-butyl-L-glutamate) (PtBuLG) and PMLGSLG (with 30 mol % of stearyl substituents), with the *tert*-butyl group as a mild acid-labile protecting group for the carboxylic acid. PtBuLG-*b*-PMLGSLG was synthesized employing the primary amine-initiated stepwise ring-opening polymerization of α -amino acid *N*-carboxyanhydrides (NCAs) in chloroform at 0 °C. The molecular weights of PtBuLG-*b*-PMLGSLGs were characterized by ¹H NMR (CDCl₃) and gel permeation chromatography (tetrahydrofuran eluent, polystyrene standard, universal calibration).^{30,33} The *tert*-butyl group was removed using trifluoroacetic acid (TFA). PMLGSLG (DP = 118) was prepared by NCA random copolymerization in chloroform at 0 °C using triethylamine as initiator.

Langmuir–Blodgett (LB). The substrate cleaning procedure was described elsewhere.³⁰ The measurements of surface pressure–area (π -*A*) isotherms and LB film transfer were performed using a home-modified computer-controlled Lauda Filmbalance (FW2), with an accuracy of 0.05 mN/m. The water used for the subphase was purified by reverse osmosis and subsequently

through a Milli-Q filtration system. The water had a resistance of ≥ 17 M Ω and a surface tension of about 72 mN/m. PLGA-*b*-PMLGSLGs were spread from *N*-methylpyrrolidone (NMP) (Acros, 99.5%)/chloroform (Lab-Scan, 99.5%) (3/7, v/v) solutions with 1–3% (v/v) of acetic acid (Acros, 99.5%) added, at a concentration of 0.4–0.6 mg/mL. PMLGSLG was spread from a chloroform solution at a concentration of 0.6 mg/mL. Vertical transfer of a PLGA-*b*-PMLGSLG monolayer onto a cleaned silicon substrate was done at a subphase temperature of 20 ± 0.1 °C, at down- and upstroke speeds of 100 and 10 mm min^{−1}, respectively, for CoPo-59-82 and at down- and upstroke speeds of 100 and 1 mm min^{−1} for the other diblock copolymers; because the monolayer of CoPo-59-82 forms an isotropic phase (where the α -helices are oriented isotropically around the water surface) with a large average helix tilt angle, while those of the other PLGA-*b*-PMLGSLGs exhibit a liquid crystalline-like phase,^{31,32} the different transfer upstroke speed for CoPo-59-82 could be attributed to the different molecular interactions and arrangements in the different monolayer phases. Monolayers of PLGA-*b*-PMLGSLGs were deposited onto both sides of the hydrophilic substrates during the upward stroke. Transfer ratios were around unity. A multilayer film of PMLGSLG can be deposited onto a hydrophobized silicon substrate by Y-type transfer, at down- and upstroke speeds of 3 mm min^{−1}.

The Langmuir trough used was 600 mm long and 200 mm wide. The substrate was 10 mm wide, and the total immersion length was 12 mm. The thickness of the substrate was 1000 μ m.

Removal of One-Sided Layer. The monolayer on one side of the transferred LB film can be removed by treating with a NMP/trifluoroacetic acid (7/3, v/v) mixture. After 1 h, the treated substrate side was rinsed with Milli-Q water and then with methanol, and finally was blown dry with a strong flow of nitrogen.

Transmission Fourier Transform Infrared (FT-IR). Transmission FT-IR measurements of films on double-sided polished silicon substrates were performed at a resolution of 3 cm^{−1}, under vacuum on a Bruker IFS66 V/S FT-IR spectrometer equipped with a MIR DTGS detector. A sample shuttle accessory was used for interleaved sample and background scanning. A clean silicon substrate was used as the reference. Each spectrum is an average of 40 cycles of 120 scans.

The polarized transmission FT-IR measurements were performed as described above, but with the use of an automated polarizer at a resolution of 4 cm^{−1}. The sample holder was oriented such that the IR beam was perpendicular to the surface of the double-sided polished silicon substrate supporting a polyglutamate film (Scheme 2). By means of the polarizer, the electric field vector was directed to a specific direction parallel to the substrate and at an angle to the dipping direction of the substrate (*Y* axis). Only the component of the dipole transition moment in the direction of the electric field vector in the plane of the substrate was detected. The polarization angle was varied in the range 0–180° in steps of 15°. Each spectrum is an average of 20 cycles of 120 scans.

Curve-fittings of the side chain ester (C=O stretching), acid (C=O stretching), amide I (mainly C=O stretching), and amide II (mainly C–N stretching) regions of transmission FT-IR spectra were performed using Bruker OPUS software (version 4.2). The parameters were optimized using a Levenberg–Marquardt algorithm.^{34,35}

Results and Discussion

Monolayers of the amphiphilic PLGA-*b*-PMLGSLG diblock copolymers have been shown to be stable up to ca. 50 mN/m (see Figure 1 for π -*A* isotherms). A comprehensive interpretation of the isotherms can be found elsewhere.^{30–32} By vertical deposition using the LB technique, a two-sided monolayer film of PLGA-*b*-PMLGSLG can be transferred onto a hydrophilic substrate on the upstroke. The monolayer deposition was performed at the maximum surface pressures where the monolayers are still stable

(30) Nguyen, L.-T. T.; Vorenkamp, E. J.; Daumont, C. J. M.; ten Brinke, G.; Schouten, A. J. *Polymer* **2010**, *51*, 1042.

(31) Nguyen, L.-T. T.; Ardana, A.; ten Brinke, G.; Schouten, A. J. *Langmuir* **2010**, *26*, 6515.

(32) Nguyen, L.-T. T.; Ardana, A.; Vorenkamp, E. J.; ten Brinke, G.; Schouten, A. J. *Soft Matter*, DOI:10.1039/C001163K.

(33) Temyanko, E.; Russo, P. S.; Ricks, H. *Macromolecules* **2001**, *34*, 582.

(34) Levenberg, K. *Q. Appl. Math.* **1944**, *2*, 164.

(35) Marquardt, D. *SIAM J. Appl. Math.* **1963**, *11*, 431.

Figure 1. π - A isotherms of various PLGA-*b*-PMLGSLGs on water surface at 20 °C. The compression speeds were ca. 19 cm² min⁻¹.

Scheme 2. Schematic Representation of the Experimental Setup for the LB Transfer (Top Picture) and the Polarized Transmission FT-IR Measurement (Bottom Picture) of a PLGA-*b*-PMLGSLG LB Monolayer on a Silicon Substrate, Presented Together with the Helix Coordinate System of a Single α -Helix Molecule in the Transferred Film

and transferable (10–15 mN/m below the collapse surface pressures and listed in Table 1). At such optimal transfer conditions, the α -helices are most densely packed and unidirectionally aligned with the minimum average helix tilt angles (θ , Scheme 2). The α -helical double-brush structure in the transferred monolayers (Scheme 1) has been evidenced by transmission FT-IR and small-angle X-ray reflectivity.³⁰ Scheme 2 shows the coordinate system of a helix molecule in the transferred LB film, presented in the experimental setup for the LB transfer and the polarized transmission FT-IR analysis. The average helix tilt angle (θ) was estimated from the transmission FT-IR absorption intensity ratio of the amide I and amide II bands characteristic of the α -helix structure using the method reported by Wieringa et al.³⁶ A curve-fitting procedure was performed to separate the spectral contributions to the amide bands from the PLGA and PMLGSLG blocks, and the helix tilt angle was calculated separately for the two blocks.³² The average helix tilt angle (θ) has been found to be

Table 1. Average Helix Tilt Angles Estimated by Transmission FT-IR for the PLGA and PMLGSLG Block in the PLGA-*b*-PMLGSLG LB Monolayers³²

Entry	PLGA- <i>b</i> -PMLGSLG	maximum transfer pressure (mN/m)	PLGA block θ_{PLGA} (°)	PMLGSLG block θ_{PMLGSLG} (°)
1	CoPo_63_39	40	77	58
2	CoPo_60_19	40	69	41
3	CoPo_59_82	35	77	74
4	CoPo_37_24	45	63	24
5	CoPo_50_11	40	36	17

Table 2. Orientations of the Transition Dipole Moments of the Amide Bands for an α -Helix, With Respect to the Helix Axis⁴⁰

vibration bands	α (°)
amide A	29
amide I	38
amide II	73

dependent on the transfer pressure and the diblock copolymer block lengths³² and is summarized for various PLGA-*b*-PMLGSLGs in Table 1.

Generally, the withdrawal of a substrate through the interface and transfer of the monolayer causes monolayer disturbance. This produces flow orientation effects which can induce orientation of the main chain of rigid rod molecules.³⁷ The transferred PLGA-*b*-PMLGSLG monolayers were observed to exhibit preferential azimuthal helix orientation with respect to the dipping direction (ϕ , Scheme 2) induced by the film transfer. The tilt azimuth was characterized with the use of polarized transmission FT-IR. The orientations of the amide transition dipole moments for a polypeptide α -helix, relative to the helix axis, are presented in Table 2. The transition dipole moment of the amide I and amide A vibrations are oriented almost parallel to the helix axis, while that of the amide II vibration is nearly perpendicular. Thus, a film with a random azimuthal helix orientation results in the same absorption intensity of the amide bands at different polarization angles. On the other hand, variation in the absorption intensity of these bands as a function of polarization angle reveals the mean azimuthal orientation with respect to the dipping direction as well as the azimuthal orientation order (e.g., see Figure 2). The polarization angle giving maxima in absorption intensity of the amide I and amide A bands, in accordance with a minimum in absorption intensity of the amide II band, indicates the azimuthal angle (ϕ , Scheme 2) by which the helices are tilted to the dipping direction. The degree of azimuthal orientation is characterized by an order parameter S , which can be calculated from the transmission IR dichroic ratio R using the following relations:^{38,39}

$$R = \frac{A_{\perp}}{A_{\parallel}} \quad (1)$$

$$S = \frac{(1-R)}{(2R+1)} \cdot \frac{2}{(3 \cos^2 \alpha - 1)} \quad (2)$$

where $A_{\parallel}$ and $A_{\perp}$ are the amide band absorptions at the polarization angle parallel and perpendicular to the preferential alignment direction of the helices, respectively, α is the angle between the transition dipole moment of the amide band and the helix axis

(36) Wieringa, R. H.; Siesling, E. A.; Werkman, P. J.; Angerman, H. J.; Vorenkamp, E. J.; Schouten, A. J. *Langmuir* **2001**, *17*, 6485.

(37) Malcolm, B. R. *J. Colloid Interface Sci.* **1985**, *104*, 520.

(38) Schmitt, F.-J.; Müller, M. *Thin Solid Films* **1997**, *310*, 138.

(39) Müller, M.; Kessler, B.; Lunkwitz, K. *J. Phys. Chem. B* **2003**, *107*, 8189.

Figure 2. Transmission FT-IR spectra at various polarization angles (0° – 180° in steps of 15°) for an LB monolayer of CoPo_{63_39} transferred at 40 mN/m onto a silicon substrate after film stabilization, measured along the Y axis at an immersion length of 2 mm (a), 5 mm (b), 7 mm (c), and 10 mm (d).

Figure 3. Top: amide I band absorption intensity as a function of the polarization angle derived from the spectra in Figure 2 for an LB monolayer of CoPo_{63_39} transferred at 40 mN/m onto a silicon substrate after film stabilization. Bottom: average azimuthal angle (open dots) and azimuthal order parameter (filled dots) as a function of the immersion length derived from the top figure.

(Table 2). In this work, the order parameter S was calculated from analysis of the integrated area of the amide I absorption band.

For analyzing the orientation effects in the monolayer on the front side (side facing the moving barrier) or on the back side of the substrate, the monolayer on the other side was removed by one-sided treatment using a solvent/acid mixture. The transmission FT-IR absorption intensity of the film after the treatment is half that before the treatment (see Supporting Information for transmission FT-IR spectra), indicating a successful one-sided removal of the monolayer.

Azimuthal Orientation Changes during Monolayer Transfer after Stabilization. Figure 2 shows the polarized transmission FT-IR spectra of the LB monolayer of CoPo_{63_39} transferred onto a silicon substrate after stabilization, measured at different immersion lengths on the substrate. The corresponding amide I band absorption intensity as a function of the polarization angle, the azimuthal angle, and the order parameter deduced from the spectra in Figure 2 are presented in Figure 3. There was no variation in absorption intensity of the side chain methylene stretching bands at 2925 cm^{-1} and 2855 cm^{-1} , and the $\text{C}=\text{O}$ stretching bands of the ester and acid groups at 1738 and 1711 cm^{-1} . This indicates that these groups are oriented randomly around the α -helices.

It is seen that the molecules deposited close to the top of the substrate, corresponding to an immersion length of 2 mm, are oriented perpendicular to the dipping direction ($\varphi = 90^{\circ}$). On further transfer, along the substrate the helix orientation changes toward parallel to the dipping direction ($\varphi = 0^{\circ}$), with the order parameter increasing with immersion length. At about the middle

Scheme 3. Schematic Representation of Two-Dimensional Flow Pattern Induced by LB Vertical Film Transfer in Monolayers of Hairy-Rod Polymers, with the Substrate Width Much Smaller than the Trough Width²²

of the film, a transition orientation at $\varphi = 60^\circ$ is observed, however, with a very small degree of order. The change in the azimuthal orientation of the helices in the transferred film along the vertical direction of the substrate can be explained by the flow orientation and deformation of the monolayer during transfer. A convergent flow produced by vertical dipping and withdrawal of the substrate through the interface, as shown in Scheme 3, brings about the orientation of the main chain axis toward the dipping direction. This two-dimensional flow pattern, induced by a two-sided vertical film transfer in a transfer geometry where the substrate is much smaller in width than the trough, has been reported by Schwiegl et al.²² for monolayers of hairy-rod polymers, including the hydrophobic PMLGSLG. Whereas for PMLGSLG the molecules are randomly oriented in the compressed Langmuir monolayer⁴¹ as well as in the first transferred area of a single LB monolayer,²² in-plane anisotropy seems to already be formed in the compressed monolayer of PLGA-*b*-PMLGSLG. The lateral molecular orientation parallel to the barrier (perpendicular to the dipping direction) is probably generated during film compression and stabilization. It was also noted that, compared to PMLGSLG, the stabilization time of the PLGA-*b*-PMLGSLG monolayers, in the surface pressure range 20–45 mN/m, was much longer, most likely due to rearrangement of the molecules until the equilibrium state was achieved. Our surface potential measurements recorded for the PLGA-*b*-PMLGSLG monolayers have indicated the gradual formation of unidirectional aligned α -helices upon monolayer compression.³¹ Since the compression process of the monolayers was accompanied by a significant increase in chain density and increase in chain tilting order, a preferential main chain alignment was probably favored during the process.

Helix Azimuth Induced by Flow Orientation. The azimuthal helix orientation and azimuthal order in the transferred PLGA-*b*-PMLGSLG monolayers were studied as a function of relative position of dipping with respect to the previous transfer position and immersion length.

Figure 4 shows the amide I band absorption intensity as a function of polarization angle and the azimuthal order for an LB monolayer of CoPo_{63_39} transferred onto a silicon substrate with the dipping position the same as for the previous transfer (see Supporting Information for polarized transmission FT-IR spectra). Originating from both the flow pattern retained in the monolayer generated by the previous transfer and the convergent flow effect caused by the transfer itself, the helices are oriented parallel to the dipping direction with a high order parameter S of 0.7. As shown in Figure 4, the order parameter increases negligibly with immersion length. Furthermore, a successive monolayer film transfer resulted in the same azimuthal order. This suggests that, after the first transfer with

Figure 4. Amide I band absorption intensity as a function of the polarization angle (top figure) and the azimuthal order parameter as a function of the immersion length (bottom figure) for an LB monolayer of CoPo_{63_39} transferred at 40 mN/m onto a silicon substrate at the same dipping position as for the previous transfer.

an immersion length of about 10–12 mm, the azimuthal orientation order (developed at the middle of the convergent flow region) almost reaches a stationary value. The azimuthal order in the back-side monolayer is somewhat smaller than that in the front-side film.

Peculiarly, after a transfer, e.g., performed at the middle of the trough (position 1, Scheme 2), for the next transfer when moving the dipping position along the X axis to a position at a distance of 25 mm away from the previous one (position 2, Scheme 2), the transferred monolayer film of PLGA-*b*-PMLGSLGs exhibited a strong azimuthal orientation at an angle of 30–60°. The amide I band absorption intensity as a function of polarization angle and the azimuthal order parameter are shown in Figure 5 for a monolayer film of CoPo_{63_39} with the transfer condition as just described (see Supporting Information for representative polarized transmission FT-IR spectra). The spectra indicate a φ of 45° with a high order parameter $S \approx 0.5$. Measurements at various positions along the horizontal direction (X axis) in the transferred film gave the same results. The influence of dipping position relative to the previous one on the azimuthal helix orientation is summarized in Table 3. Experiments with the dipping position at other distances in the range 20–30 mm from the previous transfer position also showed a preferential orientation with φ equal to 30–60° but with less order. A successive transfer after the one having this effect (entry 4, Table 3) or dipping at a distance further from the previous transfer (entry 5, Table 3) resulted in an azimuthal orientation along the dipping direction.

The effect observed can be ascribed to the flow history in the monolayer caused by the previous transfer. As depicted in Scheme 3, there is a convergent flow, and at the two sides of the sink flow, the α -helix rods can be rotated by the shear flow and aligned at an angle relative to the compression direction. Upon the next transfer at the

(41) Kurthen, C.; Nitsch, W.; Stöckelhuber, W. *Makromol. Chem., Macromol. Symp.* **1991**, *46*, 371.

Figure 5. Amide I band absorption intensity as a function of the polarization angle (top figure); the azimuthal angle (open circles) and azimuthal order parameter (filled shapes) as a function of the immersion length (bottom figure) for an LB monolayer of CoPo_{63_39} transferred at 40 mN/m onto a silicon substrate with the dipping position (in the *X*-axis direction) at 25 mm away from that of the previous transfer.

Table 3. Influence of Dipping Position Relative to That of the Previous Transfer on the Azimuthal Helix Orientation for the LB Monolayer of CoPo_{63_39} Transferred at 40 mN/m onto a Silicon Substrate

entry	dipping position of the previous transfer	dipping position (in the <i>X</i> -axis direction)	φ (°) ^a
1	after stabilization	1 ^a	90–0
2	1 ^a	1 ^a	0
3	1 ^a	2 ^a (25 mm to the left from 1)	45
4	2 ^a	2 ^a	0
5	2 ^a	3 ^a	0
6	1 ^a	3 ^a (25 mm to the right from 1)	135

^a See Scheme 2.

side of the deformed monolayer region, the orientation of the transferred molecules was affected by both the preoriented flow pattern in the compressed monolayer and a flow effect generated by the new transfer. The latter is indicated by a slight change in the azimuthal orientation from $\varphi = 60^\circ$ to $\varphi = 45^\circ$, but with a negligible change in order with increasing immersion length (Figure 5). As seen in Figure 5, the azimuthal order parameter *S* for the front-side layer is larger than for the back-side layer. This indicates that the shear flow effect due to the transfer, accompanied by the monolayer compression, is much greater for the side facing the compression barrier. Because the interval between the two successive transfers was large (ca. 20 min), the preserved helix orientation in the subsequent transfer indicates that relaxation of the orientation is very slow, if it occurs at all. Besides, varying the transfer speed (upstroke speeds of 1 and 30 mm min^{−1}) resulted in the same azimuthal angle φ of 45°. Different from other hydrophobic rigid rod polypeptides, the

Figure 6. Average helix tilt angle of the PMLGSLG block for the LB monolayers of PLGA-*b*-PMLGSLGs transferred onto silicon substrates at 35–45 mN/m (see Table 1) and the azimuthal order parameter for the LB monolayers of PLGA-*b*-PMLGSLGs with the transfer condition as entry 2 (Table 3) ($\varphi = 0^\circ$) and with the transfer condition as entry 3 (Table 3) ($\varphi = 45^\circ$). For comparison, the helix tilt angle and azimuthal order of a multilayer LB film of PMLGSLG transferred at 20 mN/m onto a hydrophobized silicon substrate (4 layers on each side) is also presented.

molecular arrangement in the monolayer flow pattern of PLGA-*b*-PMLGSLGs can be relatively stably maintained throughout the next transfer, in spite of a new convergent flow produced tending to orient the rods toward the dipping direction.

The transfer-induced azimuthal orientation phenomenon with $\varphi = 45^\circ$, only observed for the amphiphilic PLGA-*b*-PMLGSLGs, can probably be related to the molecular organization characteristic of their highly packed double-brush structure. The induced orientation phenomenon studied here affected only the tilt azimuth and not the tilt order. Brewster angle microscopy images of the PLGA-*b*-PMLGSLG monolayers upon compression to high surface pressures showed homogeneous films with no formation of any macroscopic domains.³⁰ Moreover, it has been implied that at the transfer pressures investigated (Table 1) in the monolayer of CoPo_{59_82} the PMLGSLG block segments are oriented isotropically around the water surface normal with an average tilt angle, whereas the monolayers of the other diblock copolymers might exhibit a smectic C-like phase.³² Therefore, the same transfer-induced orientation phenomenon observed for all PLGA-*b*-PMLGSLGs strongly suggests the orientation of individual polymer chains. The individual arrangement of a helix is in a strong relation with the surrounding ones as a whole. Besides, the side chain mantle around the α -helices of the PMLGSLG block provides the molecular mobility at the interface. As a result, the molecules can be strongly sheared toward a preferential orientation by any monolayer deformation caused by the combined effects of shear flow and compression. This azimuthal orientation can be significantly maintained because of strong mutual relations of the parallel α -helices in the double-brush.

To assess the influence of the double-brush structure on the phenomenon, the same experiments on the dipping-induced azimuthal orientation were performed for other PLGA-*b*-PMLGSLG diblock copolymers with different block lengths and tilt orders (Table 1). The influence of the PMLGSLG and PLGA blocks can be indicated separately by studying the azimuthal orientation effect in the LB films of, respectively, PMLGSLG and a PLGA-*b*-PMLGSLG diblock copolymer with the PMLGSLG block oriented nearly perpendicular to the interface. The results for the azimuthal orders obtained are shown in Figure 6. Because the chain

density at the interface and the average helix tilt angle of the double-brush monolayers are determined by the PMLGSLG hydrophobic block length,³² it appears that the PMLGSLG block also has a crucial role on the transfer-induced azimuthal helix orientation. The first series of experiments corresponds to the same dipping position as for the previous transfer (entry 2, Table 3), resulting in an azimuthal helix alignment along the dipping direction. The results showed that PLGA-*b*-PMLGSLGs with the PMLGSLG block tilt angle θ in the range 40–75° had high order parameters S of 0.7–0.8, which are slightly lower than for a multilayer LB film of PMLGSLG. However, the order parameter considerably decreases with decreasing the tilt angle of the PMLGSLG block. For CoPo_50_11 consisting of a very small PMLGSLG block with a nearly perpendicular helix orientation, the azimuthal order parameter S is even lower than 0.1.

On the other hand, a similar trend for the effect of PMLGSLG block on the transfer-induced orientation was also found in the experiments with the transfer position at the side of the convergent flow generated by the previous transfer (entry 3, Table 3). As indicated in Figure 6, the azimuthal orientation tilt from the dipping direction by an angle $\varphi = 45^\circ$ was only observed in the transferred films of PLGA-*b*-PMLGSLGs with the PMLGSLG block tilted by a sufficiently large angle θ_{PMLGSLG} with respect to the surface normal. When the α -helices of the PMLGSLG block are oriented nearly perpendicular to the surface, for CoPo_50_11, the molecules were azimuthally oriented parallel to the dipping direction regardless of the dipping position, but still with an order parameter not exceeding 0.1. Furthermore, the azimuthal order found in these experiments correlates increasingly with the average helix tilt angle of the PMLGSLG block. Up to $\theta_{\text{PMLGSLG}} = 74^\circ$, the order parameter almost reaches the optimal value often found for the in-plane helix orientation of hairy-rod polymers.^{22,38}

Therefore, it can be concluded that the azimuthal helix orientation phenomena induced by the transfer process in the LB monolayers of PLGA-*b*-PMLGSLGs arise mainly from the

molecular shear of the PMLGSLG hydrophobic block. The observed effects are derived from the rod-like feature and especially from the parallel helix packing and the helix tilting at the interface specific for the double-brush structure. Irrespective of the fact that the PMLGSLG block segments are oriented either isotropically or preferentially in the stabilized monolayers, they can be azimuthally oriented strongly under the influence of the flow pattern induced by the substrate dipping.

Conclusions

The influence of dipping position relative to the previous transfer on the azimuthal helix orientation has been found in the LB monolayers of the amphiphilic PLGA-*b*-PMLGSLG diblock copolypeptides. The dipping position can be manipulated resulting in an azimuthal alignment of the α -helices in the transferred films parallel to or tilted by an angle of 45° with respect to the dipping direction. The azimuthal anisotropy originates from the flow history retained in the compressed monolayer and arises mainly from deformation of the PMLGSLG top brush layer upon transfer. Consequently, the azimuthal order of the transferred PLGA-*b*-PMLGSLG LB monolayers was low for the diblock copolymers with the PMLGSLG block oriented closely to the surface normal and increased with increasing tilt angle between this block and the substrate normal. For the diblock copolymers with θ_{PMLGSLG} in the range 40–75°, significant azimuthal orientations, indicated by high order parameters S of 0.5–0.75, were obtained.

Acknowledgment. This research is supported by NanoNed, a nanotechnology program of the Dutch Ministry of Economic Affairs.

Supporting Information Available: Transmission FT-IR spectra. This material is available free of charge via the Internet at <http://pubs.acs.org>.