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Patterns of Diacetylene-Containing Peptide Amphiphiles Using Polarization Holography

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Abstract: The polarization dependence of a diacetylene polymerization was studied. For this purpose, peptide amphiphile fibers with a diacetylene moiety, which could only polymerize in the direction of the fiber, were employed. If nonaligned samples were illuminated with polarized light, only the fibers parallel to the polarization direction of the light were polymerized. With magnetically aligned fibers, spatially selective polymerization was accomplished using polarization holography.

Introduction

The photochemical polymerization of diacetylene-functionalized molecules has been studied for years. In particular, the topochemical character of this polymerization and the properties of the resulting polymer (for example, its strong absorption in the visible spectrum) have intrigued many scientists. Although diacetylene-functional materials have to be highly structured or even crystalline to allow polymerization to take place, no examples are described in the literature yet in which this level of organizational perfection is utilized to obtain control over the directionality of polymerization. Here, we show that in an isotropic sample anisotropic properties can be introduced using only the polarization of the light employed for the polymerization process. To this end, we employ peptide amphiphiles, which assemble highly controlled into straight fibers. Illuminating nonaligned samples of these fibers using plane-polarized light results in a dichroically colored, partly polymerized sample, in which only fibers parallel to the polarization direction are polymerized. When this concept of polarization-selective polymerization is applied on an aligned sample using polarization holography, patterning by spatially confined polymerization is achieved, allowing maskless lithography of diacetylenes.

Peptide amphiphiles (PAs) have gained much interest as molecular building blocks for a new generation of materials with a high degree of order on a nanoscopic level,[1] which can be applied as functional scaffolds in, for example, biomineralization[2] or cell proliferation processes.[3] In our group, we have extensively studied an eight-amino acid peptide moiety, GANPNAAG, derived from the CS protein of the malaria parasite *Plasmodium falciparum*,[4] which, when coupled to a hydrophobic tail, yields a peptide amphiphile. The tail provides a driving force for the assembly, which is directed by the hydrogen-bond forming peptide. The PAs self-assemble in water to yield fibers of which the stability can be tuned by changing the length of the hydrophobic tail.[5] In line with similar work performed by Stupp[6] and Tirrell,[7] GANPNAAG-based diacetylene-functional PAs were recently developed to make them susceptible to polymerization.[8,9] When a diacetylene moiety was included in the hydrophobic tail (Figure 1a), the fibers could be polymerized using the topochemical polymerization reaction of diacetylenes[10] (Figure 1b), indicating the well-defined packing of the diacetylenes within the fiber.[9] The PA fibers could also be aligned using a magnetic field and polymerized subsequently.[8] From the pronounced dichroic behavior of the resulting polymer, the chromophoric centra of the polymeric backbone were shown to be aligned parallel to the magnetic field.

Results and Discussion

Introducing Anisotropy. The combination of these properties, fibers with tunable stability, which can be aligned and polymerized, subsequently yielding aligned chromophoric centra, led us to hypothesize that also the polymerization-initiating event, the absorption of light by the monomer, might be direction dependent. If so, polymerization would be initiated faster for light that has the more strongly absorbed direction of polarization, subsequently leading to a polarization-dependent rate of polymerization. To investigate this possibility, nonaligned

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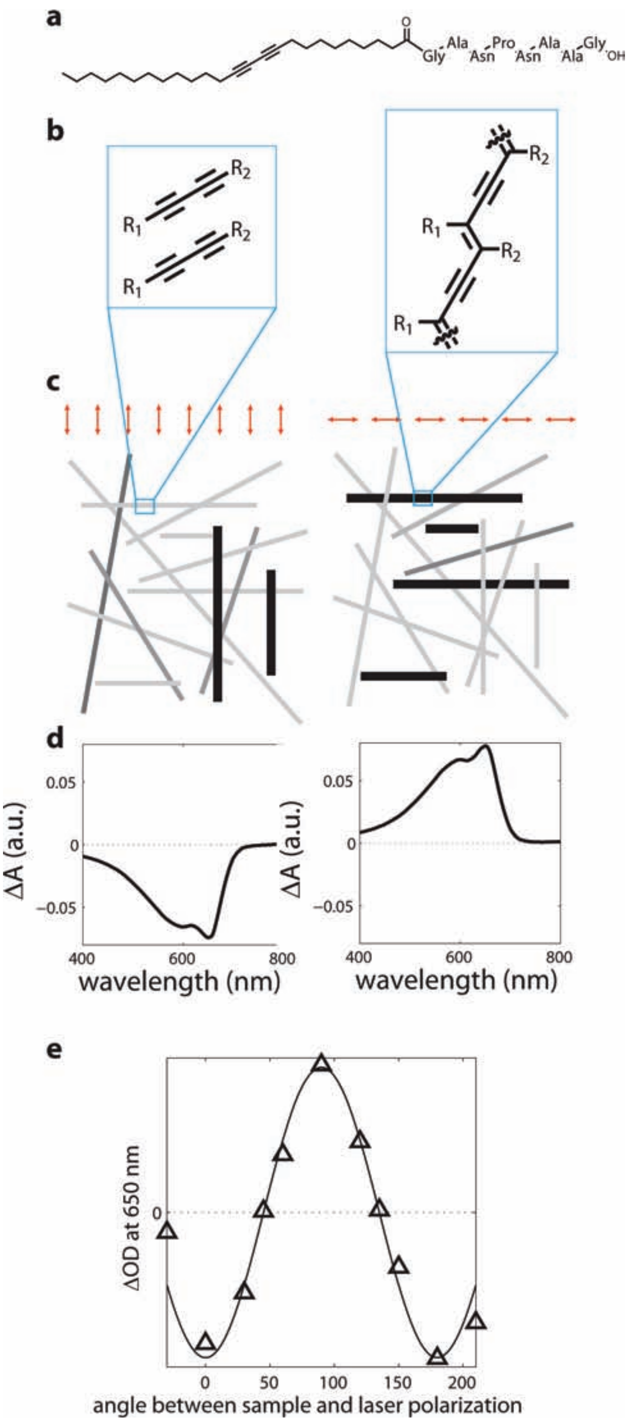

Figure 1. Illumination of a nonaligned sample with polarized light. (a) The peptide amphiphile used for the polarization-sensitive polymerization experiments. (b) The polymerization that occurs upon illumination of a diacetylene. (c) Dependent on the polarization direction of the light (the arrows), one or the other orientation of the fibers will polymerize. The darkness of the lines depicts their degree of polymerization. A black line is fully polymerized, and light gray is nonpolymerized. (d) In agreement with this theory, vertically polymerized yields indeed a negative linear dichroism (LD) signal (left), defined as $\Delta A = A_{\parallel} - A_{\perp}$. Horizontally polymerized fibers yield a positive LD signal (right). (e) The determined angle dependence of the polymerization process, monitored as the LD signal at 650 nm.

samples were polymerized with plane polarized 457 nm laser light. After polymerization, anisotropy of the samples was investigated using linear dichroism spectroscopy (LD). In Figure 1c is shown the expected polymerization for horizontally and vertically polarized light, and in Figure 1d are shown the corresponding measured LD spectra.

Light parallel to the fiber was expected to be absorbed and thus induce polymerization, where light perpendicular to the sample yields no or negligible polymerization, if the rate of polymerization is indeed polarization dependent. This would result in dichroic behavior of the resulting, partly polymerized material. Figure 1e shows the observed linear dichroism signal at 650 nm of the polymerized sample plotted against the angle under which the sample was polymerized. A negative signal indicates, as shown in Figure 1d, a vertical orientation of the polymers, while a positive signal shows a horizontal orientation. From this experiment, it can be concluded that indeed the fibers show pronounced dichroic behavior resulting from the dependence of the rate of polymerization on the polarization of the incident light. As a consequence, anisotropic properties are installed in an otherwise isotropic sample.

Patterning Using Polarization Holography. Because of this polarization-dependent rate of polymerization, we furthermore expected patterning of aligned PA fibers to be possible using polarization holography. Of the different methods described to create micro- to nanometer-sized patterns in polymeric films, holography has a special feature in that no template, mask, or mechanical contact with the film is required. The pattern is induced by a periodical variation of the intensity of the incident light onto a sample. The difference in intensity causes a difference in response, which, in its turn, leads to pattern formation. Polarization holography is a variation of this technique, in which not the intensity but the polarization direction of the light is modulated. This is accomplished by combining two circularly polarized laser beams with opposite handedness (Figure 2a).[11] At the interference plane, a pattern of linearly polarized light is obtained.

Polarization holography has been used before to create patterns in polymeric films in several ways. In one approach, a dichroic photoinitiator was employed, which dissociates preferentially with light of one polarization, initiating the polymerization at that specific spot.[12] In a second method, polymers or linear photopolymerizable polymers were used, which could reorient themselves toward the polarization of the incident light,[13−16] creating a fully polymerized sample of which the orientation of the monomers followed the polarization pattern. In all of these methods, the orientational freedom of the monomers causes the pattern to form. To our knowledge, patterning a solid film with polarization holography when the monomers do not have orientational freedom has not been reported before. This requires a unique set of characteristics of the material of which the film is composed, because the molecules have to be both prealigned and sensitive to the polarization of the incident light, so that only one polarization direction is able to initiate polymerization. Because of the alignment capabilities of the PA fibers in combination with their

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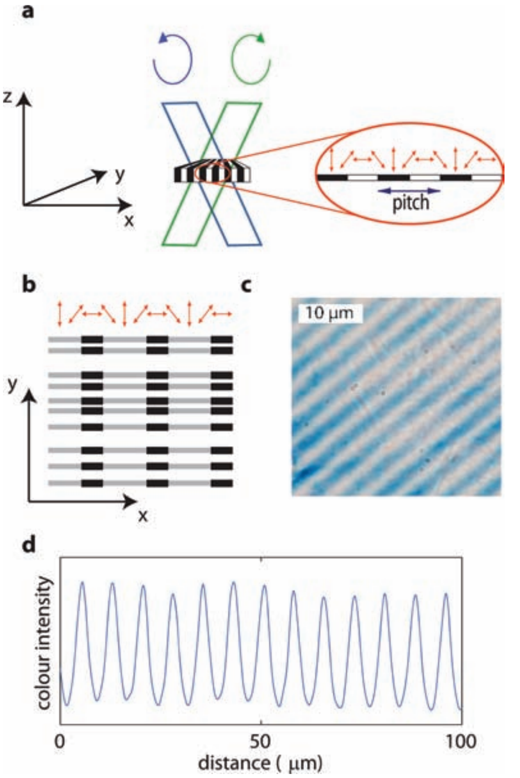

Figure 2. Setup and expected experimental results of polarization holography. (a) Schematic representation of the polarization holography setup. Two oppositely circularly polarized laser beams interfere to yield a linearly polarized pattern. (b) If the polymerization is polarization dependent, polymerization of an aligned sample is expected to only take place where the polarization is parallel to the fiber orientation, yielding a pattern of polymer. (c) The measured pattern of an aligned sample after illumination using polarization holography. (d) The intensity variation in the picture shown in Figure 4b, perpendicular to the blue lines.

unique susceptibility to the polarization direction of the incident light (Figure 2b), these architectures represent an ideal candidate for patterning using polarization holography. Although diacetylenes have been used for patterning before,[17−20] to our knowledge, this has never been accomplished with maskless lithographic patterning, employing the unique polarization-dependent selective polymerization.

To test this patterning technique, we prepared samples of the diacetylene-containing peptide amphiphilic fibers that were aligned in a strong magnetic field.[8] After drying, the samples were exposed to a pattern of linearly polarized light, using a polarization holography setup (Figure 2a). The result, depicted in Figure 2c, shows that patterning of diacetylene PAs using polarization holography is not only possible but even results in a very distinct pattern. The observed on−off behavior could be caused by an intensity threshold below which no polymerization takes place.

Varying the Pitch of the Polarization Holography Pattern. Polarization holography allows the pitch of the pattern to be easily varied by changing the angle between the incident circularly polarized beams. We used several pitches, varying from 1.5 μm (Figure 3a) to 5.2 μm (Figure 3b), which should depend on the angle as follows:

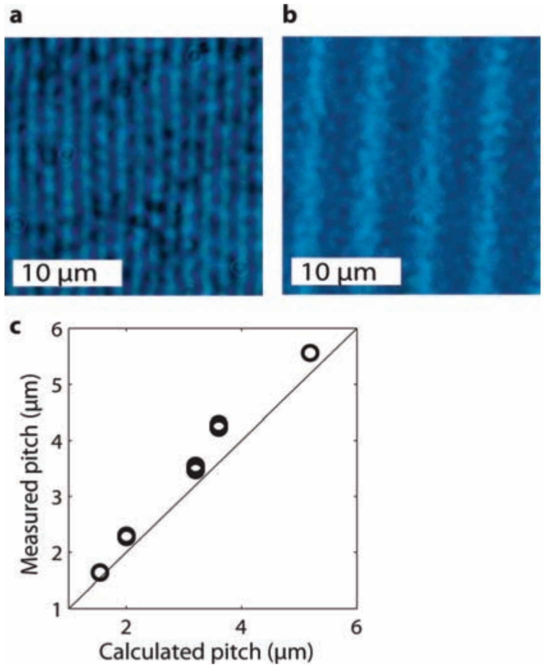

Figure 3. Varying the pitch of the polarization holography setup results in a change in the observed pattern. (a) Pitch of 1.5 μm, (b) pitch of 5.2 μm, observed with optical microscopy. (c) The pitch of the polymer patterns of several samples as compared to the calculated pitch.

$$\Lambda = \frac{\lambda}{2\,\sin(\frac{\theta}{2})}$$

where Λ is the pitch, λ is the wavelength of the incident light, and θ is the angle between the two interfering beams. The observed pitches correlated closely to the calculated ones (Figure 3c), confirming that the polymerized lines are indeed caused by the pattern of polarized light. The measured pitch was on average 10% higher than the calculated one. This may be caused by a deviation of the ideal 90° angle between the beams from the beam splitters in the setup. The angle between the beams at the sample position will then also be slightly different from the calculated angle, causing a systematic error. Furthermore, the refractive index was neglected in the calculation of the pitch and may thus also cause a small deviation.

Presence of a Threshold Value for Polymerization. Because in polarization holography the change between parallel and perpendicular light is gradual, which means that the component parallel to the fibers gradually decreases, a gradual transition between polymerized and nonpolymerized parts of the sample was expected. That would result in a gradual color change and not the sharp lines observed. The well-defined pattern observed (Figure 2c) could be caused by an intensity threshold for polymerization causing an on/off switch for polymerization. To investigate if such an intensity threshold was present, we performed polymerization experiments at UV light intensities varying from 4 J/cm^2 to less than 30 mJ/cm^2, corresponding to sample-lamp distances between 1.5 and 60 cm. The rate of polymerization had a linear dependence with the intensity of the incident light for intensities smaller than 0.6 J/cm^2, while at higher intensities the speed was lower than it would be following this relation (Figure 4). At the highest intensities (>25

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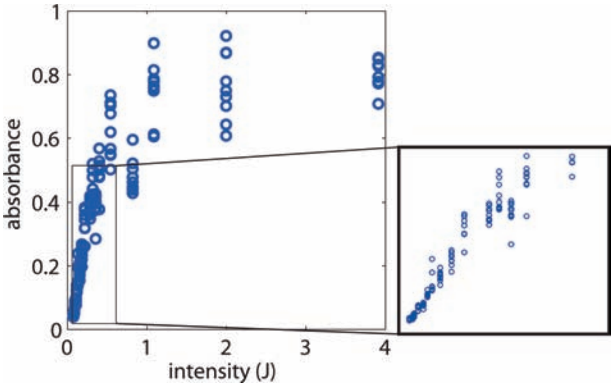

Figure 4. The absorbance of the formed polymer follows a linear relationship with the intensity of illumination for low intensity, which reaches a plateau for higher intensities.

mW/cm^2) and longer illumination times (>15 min, resulting in >22.5 J/cm^2), the position of the maximum absorbance peak shifted from 645 to 655 nm.

From these experiments, it was concluded that a threshold value, if present, seems to be even lower than 30 mJ/cm^2. The fact that no intensity threshold was measured could be due to the difference in conditions used for the experiments. First, the threshold measurements have been performed in solution, and the presence of a threshold may be influenced by the presence of oxygen. Moreover, the wavelength of the incident light may be very important for these measurements. The polarization holography was performed at 351 nm, the selective polymerization of a nonaligned sample was done at 457 nm, while the threshold measurements were performed using lower wavelength UV light, down to 254 nm. The polymerization is very sensitive toward the low-wavelength UV light, while 457 but also 351 nm are well outside the absorption band of the monomer. The intensity threshold was not investigated on dried samples using laser light of higher wavelengths because the analyses of the absorption of the sample are in that case highly dependent on the spot on the sample where the measurement takes place, yielding highly irreproducible results.

Domain Formation in Dried Samples. Remarkably, it was found that illumination using polarization holography resulted in patterns even when samples were used that had not been aligned in a magnetic field. Apparently, upon drying, some domain formation is taking place, resulting in local alignment, which is pronounced enough to yield visible differences between polymerization with parallel or perpendicular polarized light. The formed domains were large enough for the pattern to form, but did not cover the whole sample, as confirmed with linear dichroism, with which a different orientation of the fiber at different positions on the sample was measured.

To exclude patterning by intensity variation, it was verified that the intensities of the incident light did not deviate more than 1% and their ellipticities were less than 10%. Furthermore, two control experiments were performed, one on a lyophilized and hence truly nonoriented sample and a second on the nonfiber forming 10,12-pentacosadiynoic acid dried from a solution of chloroform. Both samples were spread on a glass plate and yielded a homogeneously colored sample when polymerized using polarization holography, indicating that, indeed, intensity variations cannot be the cause of the observed patterns.

Conclusions

In conclusion, we have shown that the rate of polymerization in self-assembled, diacetylene-containing PAs is dependent on the polarization of the incident light. This allowed us to employ polarized light to polymerize a randomly oriented sample to yield a dichroic colored sample, thus introducing anisotropy in an isotropic sample using only polarization of light. Furthermore, with an aligned sample and spatially addressed polarized light, polymerization takes place only at positions where the polarization of the incident light is parallel to the fiber orientation, yielding a pattern of polymer. The pitch of this pattern can easily be varied by changing the angle at which the two incident beams are combined. The possibility of using polarized light in the polymerization and hence stabilization of PA fibers opens opportunities to a further level of control within nanoarchitectures. For example, if the pattern, applied with polarization holography, is positioned perpendicular to the alignment direction of the fibers, the length of the fibers can be controlled, as is currently investigated in our group.

Experimental Section

Synthesis. The GANPNAAG peptide amphiphiles were prepared by an earlier reported procedure,[5] which is described in the Supporting Information.

Preparation of the Fiber Samples. The amphiphiles were dissolved in Milli-Q, heated to 50 °C for 30 min, followed by 15 min sonication at that temperature. Subsequently, the samples were heated to 90 °C and allowed to cool to room temperature. After this annealing, the solutions were allowed to dry on a glass substrate prior to the patterning procedure.

Sample Illumination. For the polarization holography, a 351 nm SpectraPhysics Beamlok 2085 argon ion laser with a power varying between 100 and 500 mW was used to illuminate the samples for time intervals between 1 and 10 min. The polymerization of nonaligned samples employing plane polarized light was performed with a 200 mW, 457 nm laser. LD measurements were performed on a JASCO J-810 spectropolarimeter. For the measurements to determine an intensity threshold, a Bluepoint UV lamp was employed, and UV−vis spectra were recorded on a Varian Cary-50 spectrometer.

Supporting Information Available: Experimental section describing the synthesis and characterization of the peptide amphiphiles. This material is available free of charge via the Internet at http://pubs.acs.org.

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