

Using Liquid Crystals To Image Reactants and Products of Acid–Base Reactions on Surfaces with Micrometer Resolution

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Abstract: We report that nematic liquid crystals (LCs) of 4-cyano-4'-pentylbiphenyl (5CB) and *p*-methoxybenzylidene-*p*-*n*-butylaniline (MBBA) can be used to image reactants and products of reversible, acid–base reactions on surfaces with micrometer resolution. When supported on obliquely deposited films of gold, azimuthal orientations of LCs measured on self-assembled monolayers (SAMs) formed from HOOC(CH₂)₁₀-SH are found to be orthogonal to those measured on SAMs of the corresponding sodium carboxylate salt. Conversion of the carboxylic acid to its sodium salt is amplified into a change in orientation of 10⁴–10⁵ mesogens per surface-confined acid group. The change in orientation of the LC is easily transduced into an optical output by forming twist distortions within the LC upon conversion of the carboxylic acid to its sodium carboxylate salt. The threshold conversion of acid to salt that drives the formation of the twist distortion within the supported LC can be systematically shifted between 8% and 100% by using mixed SAMs formed from HOOC(CH₂)₁₀SH and H₃C(CH₂)₁₁SH, and by pretreating these SAMs in aqueous solutions buffered between pH 1.7 and 14. We also demonstrate that region-specific conversion of the carboxylic acid to its sodium carboxylate salt can be readily imaged on micrometer scales by using LCs. Observations of the boundary region between acid and salt show no evidence of lateral transport of products and reactants over distances of micrometers over periods of days. We conclude that images of the reactants and products of this surface-confined chemical transformation can be conveniently recorded in the orientations of supported LCs.

1. Introduction

We report the use of thermotropic liquid crystals (LCs) to image reactants and products of reversible reactions between surface-confined species and reactants dissolved in solution. We demonstrate this capability by using the surface sensitivity of nematic phases of 4-cyano-4'-pentylbiphenyl (5CB) and *p*-methoxybenzylidene-*p*-*n*-butylaniline (MBBA) to amplify and optically transduce the conversion of immobilized carboxylic acid groups into their sodium carboxylate salts with a spatial resolution better than 10 μm. Whereas past studies have demonstrated the imaging of spatially resolved chemical species on surfaces by using, for example, fluorescent,¹ chemiluminescent (and electrochemiluminescent),² radioactive,³ or electroactive labels,⁴ the principles we report do not require the use of labels, are simple and fast to perform, provide spatial

resolutions of micrometers, and can, we believe, be applied to the imaging of a broad range of spatially resolved chemical species on surfaces.⁵

Liquid crystalline phases, when supported on the surfaces of solid substrates, typically assume one or more orientations that are defined with respect to the symmetry of the supporting substrates.⁶ The selection of the equilibrium orientations of the LCs is largely determined by orientation-dependent forces that act between molecules within the LC phases (mesogens) and the supporting surfaces. These forces are known to be highly sensitive to the molecular-level structure of surfaces.⁶ Because molecules within LC phases can communicate their orientations over distances of 10–100 μm on time scales of hundreds of milliseconds, the influence of the structure of a surface on the orientation of near-surface mesogens can be rapidly amplified into the bulk of a LC.⁷ Furthermore, because LCs are generally birefringent, the change in orientation of the LC can be easily transduced by using polarized light. In this paper, we exploit these properties of LCs to amplify and transduce the transforma-

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tion of self-assembled monolayers (SAMs) formed from HOOC-(CH₂)₁₀SH into their corresponding sodium carboxylate salts.⁸

The principles and procedures we report in this paper build on a past report in which LCs were used to amplify and transduce specific binding of immunoglobulins (IgGs) and avidin to SAMs presenting biotin.⁹ By supporting these SAMs on obliquely deposited films of gold¹⁰ that possessed anisotropic roughness on spatial scales (~1–10 nm) similar in size to those of proteins (~5.5 nm × 5.0 nm × 4.5 nm for avidin¹¹ and ~10 nm × 14 nm × 5 nm for IgG¹²), specific binding of anti-biotin IgG or avidin to the biotin was detected optically through a change in orientation of a LC supported on the surface. Each immobilized protein was observed to reorient ~10⁵–10⁶ mesogens within the supported LC. The change in orientation of the LC, which resulted from erasure of the anisotropic roughness of the gold/SAM surface by bound protein molecules, was easily seen with the naked eye, was imaged by optical microscopy with a spatial resolution of ~10 μm, and provides the basis of a new approach for the screening of biospecific interactions at surfaces.⁹

Whereas the study described above used LCs to image the specific binding of macromolecular species to ligands hosted on surfaces, herein we report that similar experimental procedures can be used to image chemical transformations on surfaces that result in much smaller (angstrom scale) changes to the structure of a surface. We demonstrate the design of surfaces such that the transformation of a carboxylic acid to its sodium carboxylate salt leads to an easily transduced change in the orientation of a supported LC.⁸ This demonstration is significant for several reasons. First, whereas the binding affinities of avidin and anti-biotin IgG for biotin are sufficiently strong ($K_d \approx 10^{-15}$ M for avidin in bulk solution, and $K_d \approx 10^{-9}$ M for anti-biotin IgG in bulk solution)¹³ that binding is practically irreversible, the conversion of a carboxylic acid group to its sodium carboxylate salt is an easily reversed transformation. The experiments described in this paper do, therefore, establish procedures that permit LCs to be used for the imaging of species that are reversibly bound or transformed at surfaces by contact with a solvent phase. Second, we demonstrate the design of surfaces such that the orientations of LCs can be changed at specified conversions of acid to salt (between 8% and ~100%). This result suggests that LCs can be used to quantify the extent of reactions on surfaces. Third, our results reveal that contact of LC with a surface does not erase the transformation of acid

(8) Ichimura and co-workers have shown that alignment of LCs on organic thin films hosting azobenzene and coumarin groups can be changed by illumination with light (Obi, M.; Morino, S.; Ichimura, K. *Chem. Mater.* **1999**, *11*, 656–664. Ichimura, K. In *Polymers as Electrooptical and Photooptical Active Media*; Shibaev, V., Ed.; Springer: New York, 1997; pp 138–170 and references therein). Our work, in contrast, demonstrates experimental procedures that permit the use of LCs to transduce reversible chemical reactions at surfaces that are driven by contact of surfaces with solvents containing reactants. Our work also demonstrates approaches that permit extents of reactions on surfaces to be quantified by using liquid crystals.

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(10) Obliquely deposited films of gold were prepared by vapor-phase deposition of gold at a fixed angle and direction of incidence onto stationary glass substrates. Analysis of atomic force micrographs indicates the roughness of the gold film to be greatest in a direction parallel to the direction of incidence of the gold during its deposition (Skaife, J. J.; Abbott, N. L. *Chem. Mater.* **1999**, *11*, 612–623).

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to salt, nor does it smear images of the acid and salt on micrometer scales. This result is an interesting one because the LCs used in our experiments do, in fact, contain concentrations of water of tens of millimolar.

2. Materials and Methods

2.1. Materials. We synthesized HOOC(CH₂)₁₀SH by using previously published methods.¹⁴ The liquid crystal 5CB ($T_{ni} = 34.5$ °C), manufactured by BDH, was purchased from EM Industries (Hawthorne, NY). MBBA ($T_{ni} = 40$ °C) was purchased from TCI (Portland, OR). The clearing temperatures (T_{ni}) of the LCs were measured on a Leica (Leitz, Germany) heated microscope stage. H₃C(CH₂)₁₁SH, H₃C(CH₂)₁₅SH, and cyclooctane were purchased from Aldrich (Milwaukee, WI). Cyclooctane was purified by percolation through columns of neutral alumina of Brockmann Activity I (Aldrich) until it passed the Zisman test for purity.¹⁵ Glass microscope slides were purchased from Fisher (Los Angeles, CA). Titanium (99.999%) and gold (99.999%) were purchased from Advanced Materials (Spring Valley, NY).

2.2. Methods. Cleaning of Substrates. Microscope slides were cleaned sequentially in piranha (70% H₂SO₄, 30% H₂O₂) and base solutions (70% KOH, 30% H₂O₂) using nitrogen to provide agitation (1 h at 50 °C). **Warning:** Piranha solution should be handled with extreme caution; in some circumstances, most probably when it has been mixed with significant quantities of an oxidizable organic material, it has detonated unexpectedly. The slides were then rinsed thoroughly in deionized water (18.2 MΩ-cm), ethanol, and methanol and dried under a stream of nitrogen. The clean slides were stored in a vacuum oven at 110 °C. All other glassware was cleaned in piranha solution prior to use.

Oblique Deposition of Gold. Semitransparent films of gold with thicknesses of ~100 Å were deposited onto glass slides by using an electron beam evaporator manufactured by CHA Industries (Fremont, CA). The gold was deposited from a fixed direction of incidence at an angle of incidence of 50° (measured from the normal of the surface). A 20 Å layer of titanium was used to promote adhesion between the glass and the film of gold. The rate of deposition of gold and titanium was ~0.2 Å/s. The pressure in the evaporator was less than 1 × 10⁻⁶ Torr before and during each deposition. The gold source was periodically cleaned by cycling between aqua regia (70% HNO₃, 30% HCl) and piranha solutions at 50 °C (30 min in each solution). The cycle was repeated 3–4 times, rinsing between cycles in deionized water. Gold films were routinely imaged by using atomic force microscopy (AFM).¹⁰

Formation and Pretreatment of SAMs. Self-assembled monolayers (SAMs) were formed on the surfaces of obliquely deposited films of gold by immersing the films in ethanolic solutions containing either 1 mM HOOC(CH₂)₁₀SH or a mixture of HOOC(CH₂)₁₀SH and H₃C(CH₂)₁₁SH (1 mM total thiol concentration) for 1 h. After being rinsed in ethanol and dried under nitrogen, the SAMs were immersed for approximately 1 min in aqueous solutions buffered between pH 2 and 12. Upon removal from an aqueous solution, the surface of a slide was placed under a stream of nitrogen gas to displace excess solution from the surface. Unless stated otherwise, aqueous solutions were buffered by using the following acids and sodium salts: pH 1–2, 0.1 M H₃PO₄; pH 2.5–4, 1 mM NaH₂PO₄/H₃PO₄; pH 4–5, 1 mM NaO₂-CCH₃/HO₂CCH₃; pH 6–7, 1 mM Na₂HPO₄/NaH₂PO₄; pH 8–9, 1 mM Na₂CO₃/NaHCO₃; pH 9–11, 1 mM Na₂HPO₄/Na₃PO₄; pH 11.5–12, 10 mM NaOH; and pH 12–13, 0.1 M NaOH. All buffered solutions were used within 24 h of preparation.

Region-Specific Conversion of Carboxylic Acid to Sodium Carboxylate. We spatially localized the conversion of surface-confined carboxylic acid groups to sodium carboxylate salts by using one of two procedures. First, SAMs formed from HOOC(CH₂)₁₀SH were immersed completely into aqueous solutions buffered at pH 2 and then immersed halfway into aqueous solutions buffered at alkaline pH's. Second, we translated microdroplets of alkaline aqueous solutions

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suspended from the ends of capillaries across mixed SAMs formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ and $\text{H}_3\text{C}(\text{CH}_2)_{11}\text{SH}$ that were pretreated at pH 2. The glass capillaries were mounted on an X - Y translation stage. The second procedure localized the reaction to within ~ 10 - μm -wide lines on the mixed SAM.

Ellipsometry. Ellipsometric thicknesses of SAMs were measured before and after immersion of the SAMs in aqueous solutions by using a Rudolph Auto EL ellipsometer (Flanders, NJ) at a wavelength of 6320 Å and an angle of incidence of 70°. Measurements were performed using films of gold with thicknesses of 1000 Å that were deposited without a preferred direction or angle of incidence. Ellipsometric constants were measured immediately following removal of the gold films from the evaporator. The ellipsometric thicknesses of SAMs were estimated by using a three-layer model and by assuming a refractive index of 1.45 for the monolayer.¹⁶

Contact Angles. Advancing and receding contact angles were measured using a Ramé-Hart model 100 (Mountain Lakes, NJ) goniometer at room temperature. Measurements of contact angles under cyclooctane were performed inside a glass cell that was cleaned with piranha solution prior to use. Advancing and receding contact angles were measured using procedures described previously.¹⁷

Reflectance–Absorbance FTIR Spectroscopy. Infrared spectra of SAMs supported on films of gold (1000 Å in thickness) were measured using a Nicolet Protegé 460 (Madison, WI) spectrometer with a liquid-nitrogen-cooled mercury–cadmium–telluride detector and a Graseby-Specac reflectance apparatus (Smyrna, GA) set for an angle of incidence of 84°. The spectra were collected using 2000 scans with a resolution of 2 cm^{-1} . A SAM formed from deuterated hexadecanethiol was used to collect the reference spectrum.

X-ray Photoelectron Spectroscopy. X-ray photoelectron spectroscopy (Surface Science, Mountain View, CA) was used to determine the composition of mixed SAMs formed by coadsorption of $\text{H}_3\text{C}(\text{CH}_2)_{11}\text{SH}$ and $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ ^{17,18} and to determine the extent of conversion of immobilized carboxylic acid groups to their sodium carboxylate salts. We also used XPS to survey for the presence of excess salts on the surfaces of the SAMs following their immersion in buffered aqueous solutions. The chamber pressure during acquisition of all XPS spectra was $\sim 1.5 \times 10^{-9}$ Torr, and the spot size was 250 $\mu\text{m} \times 1000 \mu\text{m}$. The intensity of X-rays emitted from the Al $K\alpha$ anode varied by $\sim 2\%$ over a 24 h period. Survey scans were acquired at each location on a sample. Acquisition was performed in Resolution 4 with 50 scans centered on the O (1s), C (1s), Na (1s), and Au (4f) peaks.¹⁹ The area of the Au (4f_{5/2}, 4f_{7/2}) peaks varied by less than 4% from sample to sample. The binding energies were referenced to the Au (4f_{7/2}) peak at 84 eV and the peak separation between the Cu (2p_{3/2}) and Cu (3s) peaks (310 eV). The Cu was initially etched with an Ar plasma to remove oxides from its surface. The etching was performed at 5 kV for 2 min in 1.5×10^{-7} Torr of Ar.

The compositions of mixed SAMs formed from $\text{H}_3\text{C}(\text{CH}_2)_{11}\text{SH}$ and $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ were estimated from normalized areas of the O (1s) peaks by using a SAM formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ as a reference.¹⁸ We do not believe that a significant amount of water was adsorbed to these SAMs at 295 K under UHV conditions. Nuzzo et al.²⁰ performed temperature program desorption (TPD) experiments and reflectance–absorbance FTIR spectroscopy within a UHV chamber at 295 K and observed hydrogen bonding between the carboxylic acid groups within a SAM formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$. Hydrogen bonding between acid groups occurs when the amount of water adsorbed is small as compared to the carboxylic acid groups.

The extent of conversion of carboxylic acid groups to sodium carboxylate salts within mixed SAMs was determined from the ratios

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(18) Bain, C. D.; Whitesides, G. M. *J. Am. Chem. Soc.* **1988**, *110*, 6560–6561.

(19) The rate of damage of SAMs formed from alkanethiols by X-rays emitted from monochromatic Al $K\alpha$ sources and secondary photoelectrons is low (Laibinis, P. E.; Bain, C. D.; Whitesides, G. M. *J. Phys. Chem.* **1991**, *95*, 7017–7021).

(20) Nuzzo, R. G.; Zegarski, B. R.; Korenic, E. M.; Dubois, L. H. *J. Phys. Chem.* **1992**, *96*, 1355–1361.

of the areas of the Na (1s) peaks (after formation of the salt) and the O (1s) peaks (prior to formation of the salt).²¹ The areas of these peaks were corrected for the atomic cross section of each species. Since the stoichiometry of the sodium carboxylate salt is two oxygens for each sodium ion, the extent of conversion of carboxylic acid to sodium carboxylate was calculated as twice the ratio of the Na (1s) and O (1s) signals. The extent of reaction was determined from the Na and O signals without further correction since both Na and O are known to reside at the surface of these SAMs.^{21,22}

Orientations of LCs. We measured the orientations of nematic phases of 5CB and MBBA on SAMs by using optical cells fabricated from two films of gold, each of which supported SAMs. First, the two gold films were aligned (facing each other) such that the direction of deposition of the gold in each film was parallel. Second, the gold films were spaced apart with a thin film of Mylar or Saran Wrap (nominal thickness 2, 12, or 30 μm).²³ A drop of 5CB or MBBA, heated into its isotropic phase ($T < 45$ °C), was then drawn by capillarity into the cavity between the two surfaces of the optical cell. The cell was subsequently cooled to room temperature and the optical texture analyzed with an Olympus BX-60 polarizing light microscope (Tokyo, Japan) in transmission mode.

Measurement of Out-of-Plane Orientations of LCs. A home-built optical apparatus was used to measure the out-of-plane orientation (tilt) of 5CB within the optical cells. The optical cells were placed between cross polars, illuminated at normal incidence using a polarized He–Ne laser, and then rotated from -20° to $+20^\circ$ with respect to the normal. A plot of the intensity of light transmitted through the cell against the angle of incidence was used to estimate the tilt of the optical axis of the LC from the surface of the cell.²⁴

Measurement of In-Plane Orientations of Liquid Crystals. We measured the azimuthal orientations²⁵ of LCs supported on SAMs by using white light, crossed polarizers, a quarter-wave plate (QWP), and ~ 2 - μm -thick optical cells. The orientation of the optical axis of the LC (the director) was determined by rotating the cell between cross polarizers and observing the shift in interference colors²⁶ upon insertion of the QWP into the optical path. The in-plane orientation of the LC was defined as the one for which the insertion of the QWP resulted in the maximum positive shift in interference colors. The interference colors were interpreted by using a Michel–Levy color chart.²⁷

Karl Fischer Titrations. The concentration of water in 5CB and MBBA was measured using a commercial Mettler DL-18 titrator (Worthington, OH).²⁸ Sample sizes ranged from 0.2 to 1.0 mL, and at least four measurements were made for each LC.

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(22) XPS^{21a–c} and ATR spectroscopy^{21d,e} have been used to demonstrate that the surface of polyethylene functionalized with carboxylic acid groups can be transformed into a surface presenting carboxylate salts by immersion in aqueous or nonaqueous solutions of high pH.

(23) By measuring interference fringes formed by monochromatic light transmitted through the optical cells, we determined the thicknesses of the cells to be within 15–20% of the nominal cell thicknesses of the Mylar spacers (12 and 30 μm). Optical cells formed using 2- μm -thick Mylar were measured to have thicknesses that ranged from 2 to 4 μm .

(24) Scheffer, T. J.; Nehring, J. *J. Appl. Phys.* **1977**, *48*, 1783–1792.

(25) We define the azimuthal (in-plane) orientation of a LC by using the direction of deposition of the gold as a reference direction.

(26) Interference colors result from the transmission of particular wavelengths of light when using a white light source of illumination. The extent of transmission of a particular wavelength of light depends on the retardation, Δnd , where Δn is the birefringence and d is the thickness of the birefringent film. Light propagating through the LC will be extinguished under conditions where $\Delta nd = j\lambda$, $j = 0, 1, 2, \dots$, where λ is the wavelength of light. Interference colors are divided into orders according to the magnitude of the retardation: 0–550 nm (first-order), 550–1100 nm (second order), and 1100–1650 nm (third order).

(27) Hartshorne, N. H.; Stuart, A. *Crystals and the Polarizing Microscope*; Edward Arnold & Co.: New York, 1970.

(28) Scholz, E. *Karl Fischer Titration*; Springer-Verlag: Berlin, Germany, 1984.

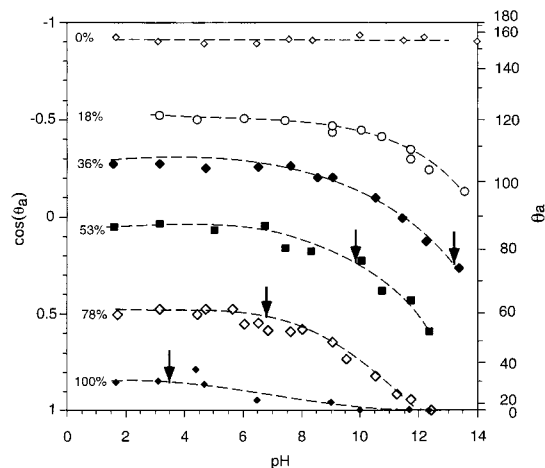


Figure 1. pH dependence of advancing contact angles of aqueous solutions measured under cyclooctane on mixed SAMs formed from $\text{H}_3\text{C}(\text{CH}_2)_{11}\text{SH}$ and $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$. The curves are labeled by the percentage of chains within the mixed SAMs that present carboxylic acid groups. The compositions of the mixed SAMs were determined by XPS (see Methods section). The solid arrows indicate the pH at which a twist distortion was observed to form within a phase of 5CB supported on the SAM.

3. Results

3.1. Characterization of Surfaces Presenting Carboxylic Acid and Sodium Carboxylate Groups. We prepared surfaces presenting carboxylic acid groups and sodium carboxylate salts by immersing SAMs formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ in aqueous solutions buffered between pH 1.7 and 13.5. Following removal of the SAMs from the aqueous solutions, bulk liquid remaining on the surfaces of the SAMs was displaced by using a stream of nitrogen gas. Because we believed it possible that processes of evaporation taking place during displacement of the aqueous solution might expose a SAM to a pH that differed substantially from the pH of the buffered solution, and because we believe it possible that evaporation might also lead to the deposition of crystals of excess salt on the surface of a SAM, we used measurements of contact angles, ellipsometry, FTIR, and XPS to characterize SAMs before and after their immersion into buffered aqueous solutions.

First, we verified that contact angles of aqueous solutions (pH 1.7–13.5) measured on SAMs formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ (under cyclooctane) were the same as contact angles reported in the past by Bain and Whitesides (curve labeled 100% in Figure 1).¹⁷ For aqueous solutions buffered to pH's greater than ~5, contact angles decreased smoothly to wetting with increasing pH.²⁹ The decrease in contact angles with increasing pH has been interpreted by others to result from ionization of immobilized carboxylic acid groups.^{17,30} Measurements based on contact angle titrations,^{17,30} the use of a quartz crystal microbalance,³¹ force–distance curves,³² and adhesion measurements³³ all indicate the onset of ionization to occur between

(29) Lee and co-workers have reported a maximum in the contact angle of water buffered at pH ~6 on SAMs formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ and $\text{H}_3\text{C}(\text{CH}_2)_{10}\text{SH}$ when the SAMs contained more than 75% $\text{HOOC}(\text{CH}_2)_{10}\text{S}$ (Lee, T. R.; Carey, R. I.; Biebuyck, H. A.; Whitesides, G. M. *Langmuir* **1994**, *10*, 741–749). Our measurements of contact angles do not display a maximum at pH ~6.

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(33) Vezenov, D. V.; Noy, A.; Lieber, C. M. *J. Am. Chem. Soc.* **1997**, *119*, 2006–2015.

pH 5 and 6. These measurements suggest that the onset of ionization of a carboxylic acid group tethered to a surface occurs at a pH higher than that for a carboxylic acid group in bulk aqueous solution ($\text{p}K_{\text{a}} = 4.7$).³⁴ We also verified that our high-resolution XPS spectra of SAMs formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ were in agreement with past measurements.³⁵

Second, by using ellipsometry and XPS, we investigated whether excess salts were deposited onto SAMs formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ upon their immersion and withdrawal from buffered aqueous solutions. By using ellipsometry, we measured the thickness of a SAM formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ prior to pretreatment with an aqueous solution to be $19 \pm 1 \text{ \AA}$.^{35a,36} Following pretreatment in aqueous solutions, the thicknesses of the SAMs were measured as follows: pH 2.5 (1 mM $\text{NaH}_2\text{PO}_4/\text{H}_3\text{PO}_4$), $23 \pm 1 \text{ \AA}$; pH 3.8 (10 μM $\text{NaH}_2\text{PO}_4/\text{H}_3\text{PO}_4$), $20 \pm 1 \text{ \AA}$; pH 4.0 (1 μM $\text{NaH}_2\text{PO}_4/\text{H}_3\text{PO}_4$), $18 \pm 1 \text{ \AA}$; pH 9 (1 mM $\text{Na}_2\text{HPO}_4/\text{Na}_3\text{PO}_4$), $20 \pm 1 \text{ \AA}$; and pH 11.7 (10 mM NaOH), $22 \pm 1 \text{ \AA}$.³⁶ Because the deposition of excess salt (or acid) onto a SAM must be accompanied by the presence of the anion of the salt (e.g., PO_4^{3-}), we used XPS to determine whether the small variations in ellipsometric thicknesses reported above were caused by the deposition of excess salt onto the SAMs. XPS signals from phosphorus were measured on the SAM immersed into the solution buffered to pH 2.5. This result and others³⁷ lead us to conclude that the increased thickness of the SAM pretreated at pH 2.5 does result from the deposition of H_3PO_4 onto the surface. In contrast, however, we measured no signal from phosphorus on SAMs formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ that were pretreated in 1 mM solutions prepared from phosphates at pH 3.2, 3.6, 6.8, 8.0, and 10.6. This result is significant because this range of pH's does, in fact, encompass solution conditions that cause changes to the orientations of LCs supported on SAMs formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ (see below). Finally, at pH 11.7, we note a small increase in the ellipsometric thickness (2–3 \AA), which likely corresponds to deposition of salts on these surfaces. At pH 11.7, a 10% decrease in the area of the Au (4f) peaks was observed in the XPS spectra, thus confirming the presence of excess salts on these SAMs.

We also used XPS to confirm that pretreatment of SAMs formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ with aqueous solutions buffered at either pH 3.2 or 10.6 resulted in the preparation of SAMs that presented COOH and $\text{COO}^- \text{Na}^+$ groups at their surfaces. Following pretreatment of the SAMs with an aqueous solution buffered at pH 3.2, the relative peak areas of an XPS spectrum were determined to be $76 \pm 1\%$ C and $24 \pm 1\%$ O, corresponding closely to the composition of the SAM prior to pretreatment in the aqueous solution ($79 \pm 2\%$ C and $21 \pm 2\%$ O). The sodium ion was not observed on the surface of SAMs formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ that were treated in aqueous solutions buffered at pH's of 3.6 or less. Thus, we conclude that the carboxylic acid group was not converted to the carboxylate salt by this procedure. In contrast, following

(34) *Handbook of Chemistry and Physics*, 67th ed.; Weast, R. C., Ed.; CRC Press: Boca Raton, FL, 1986.

(35) (a) Bain, C. D.; Troughton, E. B.; Tao, Y.; Evall, J.; Whitesides, G. M.; Nuzzo, R. G. *J. Am. Chem. Soc.* **1989**, *111*, 321–335. (b) Nuzzo, R. G.; Dubois, L. H.; Allara, D. L. *J. Am. Chem. Soc.* **1990**, *112*, 558–569.

(36) Ellipsometric thicknesses were measured to be invariant for at least 30 min in ambient air. Chidsey and Loiacono reported that atmospheric contamination caused the apparent thickness of a SAM formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ to increase when exposed to air (Chidsey, C. E. D.; Loiacono, D. N. *Langmuir* **1990**, *6*, 682–691).

(37) By using XPS, we observed the deposition of H_3PO_4 onto SAMs that were formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ and pretreated in aqueous solutions of H_3PO_4 at pH 2. Because the $\text{p}K_{\text{a}1}$ of phosphoric acid is 2.1, this acid is not fully dissociated in aqueous solution at pH 2. In contrast, when hydrochloric acid ($\text{p}K_{\text{a}} < 1$) at pH 1.8 was used, no Cl was detected on SAMs formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ by XPS.

immersion of a SAM in an aqueous solution buffered to a pH greater than 4, sodium was detected on the surface by XPS: pretreatment of the surface with aqueous solutions of increasing pH resulted in an increase in the sodium content of the surface. When the surface was pretreated at pH 10.6, the relative peak areas in the XPS spectrum of the SAM were found to be $9 \pm 2\%$ Na, $70 \pm 4\%$ C, and $21 \pm 2\%$ O. In contrast, XPS spectra of SAMs formed from $\text{HO}(\text{CH}_2)_{10}\text{SH}$ ³⁸ and pretreated with aqueous solutions buffered at pH 7.0 or 10.6 revealed the presence of negligible amounts of sodium on these surfaces. These results suggest that the sodium content of SAMs formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ and pretreated at high pH is due to the transformation of the surface-confined acid to its salt. We estimate the conversion of acid to salt on a SAM formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ when pretreated at a pH of 10.6 to be $\sim 90\%$ (see Methods section). By using XPS, we also observed the pretreatment of SAMs with aqueous solution of high pH to increase the oxygen content of these surfaces. We believe the increase in oxygen content of these surfaces was caused by hydration of the sodium carboxylate salt.

We used RA-FTIR to investigate whether the hydrocarbon regions of SAMs formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ underwent measurable reorganization upon pretreatment with the aqueous solutions. Spectra were measured before and after immersion of SAMs in aqueous solutions buffered to pH 3.2, 7.0, or 11.7. The positions of the symmetric and asymmetric CH_2 stretches in all spectra were found to be indistinguishable³⁹ before and after immersion and similar to past reports based on SAMs formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$.^{36,40} This result suggests that the hydrocarbon chains of these SAMs do not undergo substantial reorganization upon conversion of the acid to its salt.

In summary, the measurements above, when combined, lead us to conclude that experimental procedures based on the immersion and withdrawal of SAMs from buffered, aqueous solutions can be used to prepare SAMs presenting COOH and COO^-Na^+ groups at the surface of obliquely deposited films of gold. We do not believe these SAMs support excess salt in the pH range 3.2–10.6, nor do we believe that the hydrocarbon regions of the SAMs are perturbed significantly by pretreatment in the aqueous solutions.

3.2. Orientations of LCs on Surfaces Presenting Carboxylic Acid and Sodium Carboxylate Groups. We measured the orientations of LCs supported on SAMs presenting carboxylic acid or sodium carboxylate groups by assembling optical cells from two SAMs pretreated at the same pH, either 3.2 or 10.6. The thickness of the layer of LC within the cavity of the optical cell was $\sim 12 \mu\text{m}$, and the direction of deposition of gold supporting each SAM was parallel within the cell. By mounting these cells in the crystal rotation apparatus (see Methods section), we measured of the tilt of the optical axis of 5CB from

(38) Contact angles of aqueous solutions have been reported (ref 17) for SAMs formed from $\text{HO}(\text{CH}_2)_{11}\text{SH}$. When aqueous solutions buffered between pH 2 and 13 were used, the contact angle measured under cyclooctane dropped from 43° to 39° with increasing pH. The decrease in contact angle was attributed to trace impurities within the cyclooctane.

(39) We measured the positions of absorbances corresponding to asymmetric and symmetric methylene stretches (ν_a , ν_s) for SAMs formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ at pH 3.2 ($\nu_a = 2919 \text{ cm}^{-1}$, $\nu_s = 2849 \text{ cm}^{-1}$), pH 7 ($\nu_a = 2920 \text{ cm}^{-1}$, $\nu_s = 2849 \text{ cm}^{-1}$), and pH 11.7 ($\nu_a = 2920 \text{ cm}^{-1}$, $\nu_s = 2849 \text{ cm}^{-1}$). Prior to pretreatment in an aqueous solution, we measured $\nu_a = 2920 \text{ cm}^{-1}$, $\nu_s = 2850 \text{ cm}^{-1}$. These values compare favorably to past measurements of ref 40a, where $\nu_a = 2918 \text{ cm}^{-1}$, $\nu_s = 2850 \text{ cm}^{-1}$; ref 36 where $\nu_a = 2920 \text{ cm}^{-1}$, $\nu_s = 2853 \text{ cm}^{-1}$; and ref 40b,c where $\nu_a = 2919 \text{ cm}^{-1}$, $\nu_s = 2850 \text{ cm}^{-1}$.

(40) (a) Freeman T. L.; Evans, S. D.; Ulman, A. *Thin Solid Films* **1994**, *244*, 784–788. (b) Sun, L.; Crooks, R. M.; Ricco, A. J. *Langmuir* **1993**, *9*, 1775–1780. (c) Yang, H. C.; Dermody, D. L.; Xu, C.; Ricco, A. J.; Crooks, R. M. *Langmuir* **1996**, *12*, 726–735.

the plane containing the SAM to be less than 1° for cells with SAMs presenting either COOH or COO^-Na^+ groups. That is, the orientation of 5CB on these surfaces was planar,⁴¹ and no measurable change in the tilt of the LCs was evident upon conversion of the acid to its sodium carboxylate salt.

Our second set of measurements focused on the azimuthal orientation of the optical axis of 5CB and MBBA within the plane containing each SAM. First, we note that the optical textures, and thus azimuthal orientations of LCs, were observed to be uniform across all optical cells. The azimuthal orientation was determined by noting the change in interference colors upon insertion of a quarter-wave plate into the optical path (Figure 2). When cells formed with SAMs pretreated at pH 3.2 were used, 5CB and MBBA were observed to orient parallel to the direction of deposition of the gold (Figure 2A–D). In contrast, when SAMs pretreated at pH 10.6 were used, 5CB and MBBA oriented perpendicular to the direction of deposition of the gold (Figure 2E–H). A few disclination lines were typically observed within these cells. In both cases (pH 3.2 and 10.6), we observed no bulk distortion (twist, bend, or splay) of the LC within the cells.

We considered it possible that the azimuthal orientations of LCs reported above might result from deposition of excess salts (or other impurities) on the SAMs at levels not detected by XPS. Because SAMs formed from $\text{HO}(\text{CH}_2)_{11}\text{SH}$ are wet by aqueous solutions at all pH's, we investigated the orientations assumed by 5CB on these SAMs following pretreatment in solutions buffered between pH 3.0 and 11.7. Uniform alignment of 5CB in a direction parallel to the direction of deposition of the gold was observed within all optical cells pretreated between pH 3.0 and 11.7. We also formed optical cells using SAMs that were not wet by aqueous solutions (e.g., SAMs formed from $\text{H}_3\text{C}(\text{CH}_2)_{15}\text{SH}$). No change in anchoring was observed as a function of the pH of the buffer used to pretreat these SAMs. These results, when combined with the results of XPS reported in the previous section, lead us to conclude that the azimuthal orientations of LCs supported on SAMs formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ pretreated at either low pH (3.2) or high pH (10.6) differ by 90° and that this difference in orientation is caused by the conversion of the immobilized carboxylic acid group to its sodium carboxylate salt.

3.3. Enhancement of Optical Contrast by Formation of Twisted Nematic Cells upon Conversion of an Immobilized Carboxylic Acid to Its Sodium Salt. Whereas the results described in Section 3.2 establish that the azimuthal orientations of nematic LCs supported on SAMs formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ change by 90° upon conversion of the acid to its sodium salt, the changes in interference colors seen in Figure 2 are relatively subtle, and the use of a quarter-wave plate is inconvenient. In this section, we describe the design of an optical cell that provides a level of optical contrast that is sufficiently high as to be easily interpreted by using simple experimental procedures. The approach we report is based on the design of an optical cell with asymmetric surfaces such that conversion of the carboxylic acid to its sodium salt is accompanied by formation of a twist distortion within a nematic LC. Enhancement of optical contrast by using twist distortions is widely exploited in optical devices in which the state of a LC is driven between twisted and untwisted states by application of an electrical field.⁴²

(41) Evans and co-workers have reported measurements of the orientations of LCs on SAMs formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$. These measurements were not performed on obliquely deposited gold, and these authors did not investigate the influence of the conversion of acid to salt on the orientations of supported LCs (Evans, S.; Allinson, H.; Boden, N.; Henderson, J. *Faraday Discuss.* **1996**, *104*, 37–48).

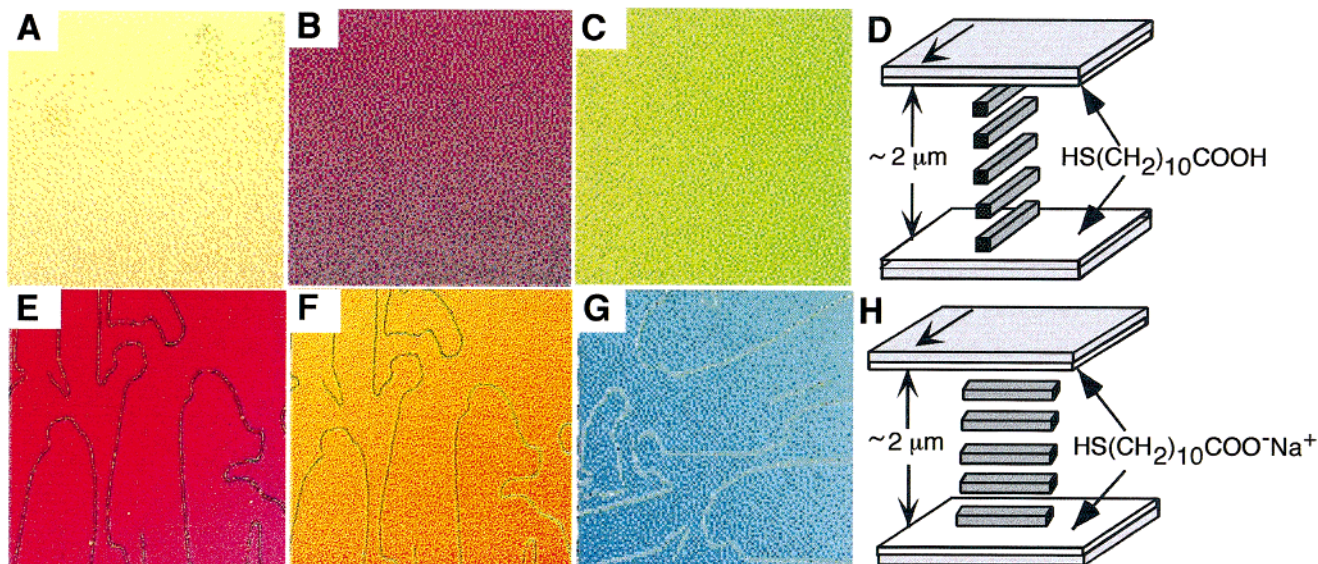


Figure 2. Interference colors and orientations of 5CB within optical cells fabricated using obliquely deposited films of gold that supported SAMs formed from HOOC(CH₂)₁₀SH. The SAMs were pretreated by aqueous solutions buffered at pH either 3.2 or pH 10.6. (A–C) pH 3.2. (A) Interference color without a QWP in the optical path; the yellow color is a second-order interference color corresponding to a retardation of ~870 nm. (B) Interference color with the optical axis of the QWP aligned parallel to the direction of deposition of the gold; the maroon color indicates an increase in retardation to ~1000 nm. (C) Interference color with the optical axis of the QWP aligned perpendicular to the direction of deposition of the gold film; the green color indicates a decrease in retardation to ~720 nm. (D) Schematic illustration of the orientation of the LC that is consistent with the interference colors shown in A–C. The bold arrow indicates the direction of deposition of gold onto the substrate. (E–G) pH 10.6. (E) Interference color without a QWP in the optical path; the red color is a first-order interference color corresponding to a retardation of ~500 nm. (F) Interference color with the optical axis of the QWP aligned parallel to the direction of deposition of the gold; the orange color indicates a decrease in retardation to ~370 nm. (G) Interference color with the optical axis of the QWP aligned perpendicular to the direction of deposition of the gold film; the blue color indicates an increase in retardation to ~650 nm. (H) Schematic illustration of the orientation of the LC that is consistent with the interference colors shown in E–G.

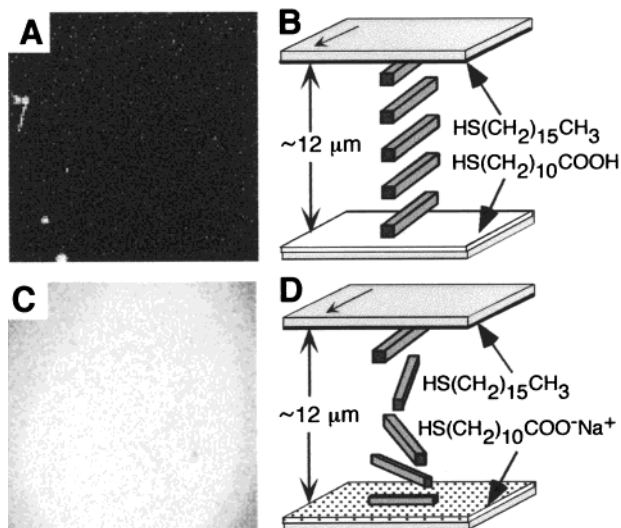


Figure 3. (A and C) Optical textures (crossed polars) formed by nematic 5CB within optical cells prepared with one surface supporting a SAM formed from H₃C(CH₂)₁₅SH and an opposing surface supporting a SAM formed from HOOC(CH₂)₁₀SH that was pretreated at pH 3.2 (A) or 10.6 (C). The bold arrows indicate the direction of deposition of gold onto the substrate. (B and D) schematic illustrations of the orientations of the LCs interpreted from the optical textures shown in A and C, respectively.

Figure 3 shows optical textures of 5CB confined to cells fabricated using a SAM formed from H₃C(CH₂)₁₅SH on one surface and a SAM formed from HOOC(CH₂)₁₀SH on the opposing surface. As shown in Figure 3A, when the SAM

formed from HOOC(CH₂)₁₀SH was pretreated at pH 3.2, the optical cell appeared dark when viewed between crossed polars. These and other optical characteristics indicate that 5CB was oriented without distortion within the optical cell (Figure 3B). When the SAM formed from HOOC(CH₂)₁₀SH was pretreated at pH 10.6, however, the cell appeared bright between crossed polars (Figure 3C) and dark between parallel polars. These optical characteristics are consistent with the presence of a twist distortion within the LC.⁴³ The presence of the twist distortion within the LC indicates that the SAM presenting COO⁻Na⁺ imposes an azimuthal orientation on the LC that is orthogonal to the orientation imposed by H₃C(CH₂)₁₅SH. This result also indicates that the strength of anchoring of 5CB on both surfaces is sufficient to induce a twist distortion in the bulk LC. We observed these twist distortions to be stable over 24 h.

3.4. Dependence of Orientation of LC on Extent of Conversion of Acid to Salt. The results presented in Section 3.3 demonstrate the design of an optical cell such that conversion of the carboxylic acid group to its sodium salt results in the formation of an easily transduced twist distortion within the LC. To determine how the orientation of the LC changes as a function of the extent of conversion of acid to salt, we measured the orientations of 5CB within cells assembled using SAMs formed from HOOC(CH₂)₁₀SH that were pretreated with aqueous solutions buffered between pH 1.7 and 12.7. Figure 4 shows the change in orientation of 5CB (formation of a twist distortion) to be almost discontinuous at pH 3.8 (Figure 4). At pH 3.8, twisted regions of 5CB with lateral dimensions of 100–1000 μm were observed to coexist with uniform (untwisted) regions of 5CB. The change from uniform to twisted alignment occurred by the growth of macroscopic ~90° twisted domains within

(42) Blinov, L. M., Chigrinov, V. G. *Electrooptic Effects in Liquid Crystal Materials*; Springer-Verlag: New York, 1994.

(43) A twist deformation of 90° rotates the polarization of light transmitted through the cell by 90°.

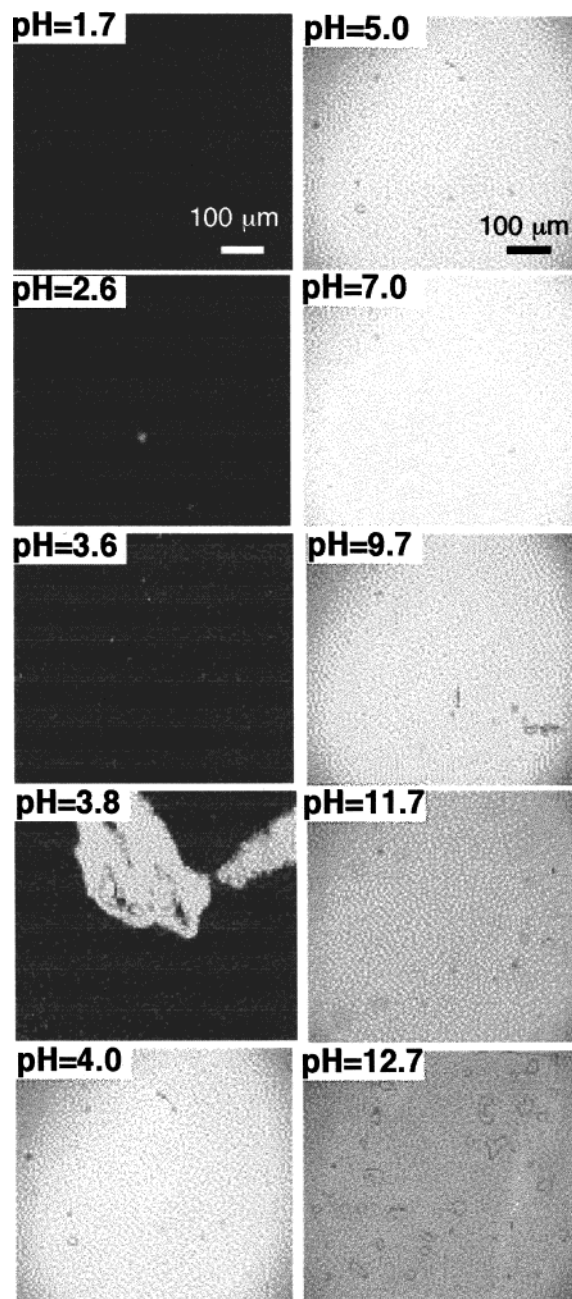


Figure 4. Optical textures (cross polars) formed by nematic phases of 5CB within optical cells fabricated with one surface supporting a SAM formed from $\text{H}_3\text{C}(\text{CH}_2)_{15}\text{SH}$ and an opposing surface supporting a SAM formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ that was pretreated with aqueous solutions buffered between pH 2 and 13. The direction of deposition of the gold was parallel to one of the polarizers.

the LC. At high pH, loop disclinations were observed in twisted areas of the cells (Figure 4). The twist angles within these loops were, in general, found to be the supplementary angle of the twist angles of the surrounding LC phase. Although we believe salts to be deposited onto surfaces pretreated at $\text{pH} < 3.2$ and $\text{pH} \geq 11.7$ (see Section 3.1), our results here indicate that the salts do not cause nonuniform orientations of 5CB.

Because a twist distortion within a LC causes plane-polarized light to rotate as it propagates through the LC,^{42,43} we found measurements of the angle of rotation of polarized light that passed through an optical cell to provide a simple and quantitative method to characterize the change in orientation of 5CB. The angle of rotation was determined by simultaneously rotating the analyzer and the optical cell with respect to the

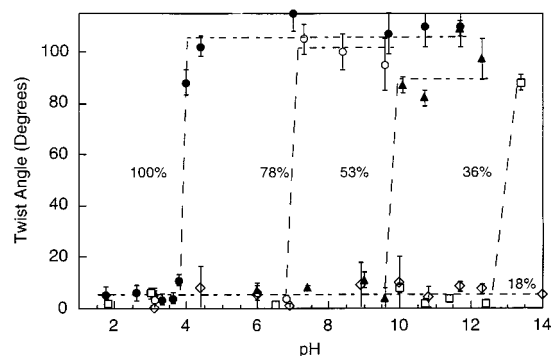


Figure 5. Angles of twist distortions within nematic phases of 5CB confined within optical cells prepared with one surface supporting a SAM formed from $\text{H}_3\text{C}(\text{CH}_2)_{15}\text{SH}$ and an opposing surface formed by coadsorption of $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ and $\text{H}_3\text{C}(\text{CH}_2)_{11}\text{SH}$ that was pretreated by immersion in aqueous solutions buffered between pH 2 and 13. The curves are labeled by the percentage of chains within the mixed SAMs that present carboxylic acid groups. The compositions of the mixed SAM were determined by XPS (see Methods section).

polarizer to obtain a minimum in the intensity of the polarized light transmitted through the sample. This angle is equal to the angle subtended by the twist distortion within the LC. The “twist angle” of 5CB measured as a function of the pH of solutions used to pretreat SAMs from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ is shown in Figure 5 (data labeled 100%). Whereas twist angles for cells pretreated with aqueous solutions with $\text{pH} < 3.8$ were $0\text{--}10^\circ$, twist angles for cells pretreated at $\text{pH} > 3.8$ ranged from 85 to 105° . The twist angles were not exactly 0° and 90° , respectively, for two reasons. First, the alignment of the glass slides during the evaporation of gold films was controlled only to within $\sim 10^\circ$. Second, when aligning the two opposing SAMs during fabrication of the optical cells, we found errors of $\sim 5^\circ$ to be common.

3.5. Design of Surfaces That Trigger Changes in Orientation of 5CB at Specified Conversions of Acid to Salt. The results described above demonstrate that the azimuthal orientations of 5CB on SAMs formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ change discontinuously as a function of the extent of conversion of acid to salt. Because the discontinuous transition makes possible determination of a threshold conversion of acid to salt but provides no dynamic range to the measurements, here we illustrate one approach that permits the orientation of 5CB to be triggered at different threshold conversions of acid to salt. The approach is based on the preparation of mixed SAMs formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ and $\text{H}_3\text{C}(\text{CH}_2)_{11}\text{SH}$. We formed mixed SAMs using $\text{H}_3\text{C}(\text{CH}_2)_{11}\text{SH}$ because we previously measured $\text{H}_3\text{C}(\text{CH}_2)_{11}\text{SH}$ to align 5CB (and MBBA) parallel to the direction of deposition of the gold.⁴⁴ An increase in the mole fraction of $\text{H}_3\text{C}(\text{CH}_2)_{11}\text{SH}$ within mixed SAMs formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ and $\text{H}_3\text{C}(\text{CH}_2)_{11}\text{SH}$ would, we hypothesized, increase the extent of conversion of acid to salt necessary to trigger a change in the azimuthal orientation of 5CB.

We prepared LC cells by forming a SAM from $\text{H}_3\text{C}(\text{CH}_2)_{15}\text{SH}$ on one surface of the cell and a mixed SAM from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ and $\text{H}_3\text{C}(\text{CH}_2)_{11}\text{SH}$ on the opposing surface of the cell. Prior to assembly of the cell, the entire surface of the mixed SAM was pretreated with a buffered aqueous solution. Whereas pretreatment of a SAM formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ with an aqueous solution buffered at pH 3.8 induced a twist distortion within a nematic phase of 5CB, a mixed SAM containing a mole fraction of acid groups (X_{COOH})⁴⁵ of 0.36 (as determined

(44) Gupta, V. K.; Abbott, N. L. *Phys. Rev. E* **1996**, *54*, R4540–R4543.

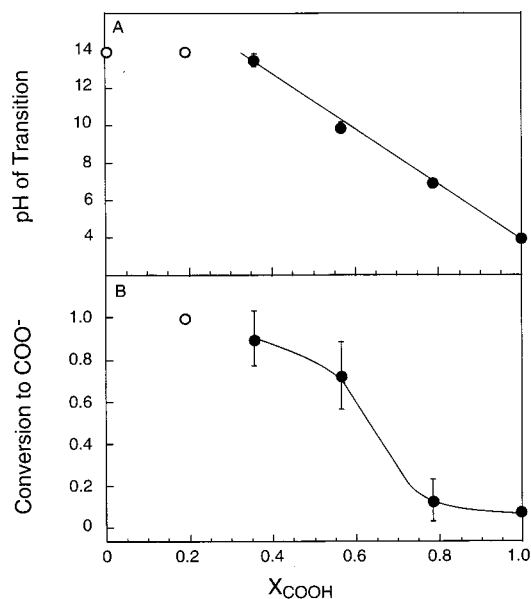


Figure 6. (A) pH of aqueous solutions that triggered twist distortions within nematic phases of 5CB supported on mixed SAMs formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ and $\text{H}_3\text{C}(\text{CH}_2)_{11}\text{SH}$. The compositions of the mixed SAMs (X_{COOH}) were determined by XPS. (B) Extent of conversion of immobilized carboxylic acid groups to sodium carboxylate salts at onset of twist distortions of 5CB as a function of the composition of mixed SAMs formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ and $\text{H}_3\text{C}(\text{CH}_2)_{11}\text{SH}$. Filled data points correspond to mixed SAMs that caused twist distortions, and open data points correspond to mixed SAMs that did not cause twist distortions of the LC.

by XPS) did not cause the formation of a twist distortion within 5CB until the pH of the solution approached 13.5 (Figure 5). Mixed SAMs containing $X_{\text{COOH}} = 0.78$ and $X_{\text{COOH}} = 0.53$ caused twist distortions to form in 5CB at threshold pH's of 7.0 and 10.0, respectively. These results are summarized in Figure 6A, which shows the pH of the buffer that caused twist distortions to form within 5CB as a function of the mole fraction of acid within the mixed SAM. A linear relation between the threshold pH and composition of the SAM is observed for SAMs with compositions $0.36 < X_{\text{COOH}} < 1.0$. The extent of conversion of acid to salt within the mixed SAM that triggered the twist distortion of 5CB was determined by XPS (Figure 6B). Whereas conversion of ~8% of acid groups to sodium salts was sufficient to change the orientation of 5CB supported on SAMs formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$, almost complete conversion of acid to sodium salt was required to trigger a change in orientation of the LC supported on a mixed SAM with composition $X_{\text{COOH}} \approx 0.36$. Mixed SAMs with composition $X_{\text{COOH}} < 0.36$ were observed to align LCs parallel to the direction of deposition of gold at all pH's and did not induce a twist distortion within the nematic phase. This result indicates that $\text{S}(\text{CH}_2)_{11}\text{CH}_3$ controls the orientation of 5CB supported on mixed SAMs containing less than 36% carboxylic acid. Finally, we note that excess salts are not deposited onto mixed SAMs during pretreatment with aqueous solutions buffered at high pH because the receding contact angles of the aqueous solutions (in air) used to pretreat these SAMs were finite. For example, the receding contact angle of an aqueous solution buffered at pH ~13.5 was 60° on a mixed SAM with composition $X = 0.18$.

(45) X_{COOH} is defined as $N_{\text{COOH}}/(N_{\text{CH}_3} + N_{\text{COOH}})$, where N_{COOH} is the number of chains terminated with carboxylic acid groups and N_{CH_3} is the number chains terminated with methyl groups.

3.6. Imaging of Reactants and Products: Spatial Resolution and Stability of Image. The results above demonstrate that LCs can be used to report optically the extent of conversion of immobilized carboxylic acid groups to sodium carboxylate salts. Here we show that 5CB can also be used to image simultaneously both acid and salts on surfaces on which the transformation of acid to salt takes place in a spatially resolved manner. We prepared LC cells by forming a SAM from $\text{H}_3\text{C}(\text{CH}_2)_{15}\text{SH}$ on one surface and a SAM from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ on the opposing surface of the cell. Prior to assembling the cell, the entire surface of the SAM formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ was pretreated at pH 3.2. This surface was subsequently dipped halfway into an aqueous solution at pH 10.6. Figure 7A and B shows the optical textures of a cell prepared in this manner. When viewed through crossed polars (Figure 7A), region I (pH 3) appears dark due to the extinction of light transmitted through the cell by the analyzer. This result and others indicate that 5CB is anchored in region I in the same azimuthal direction on both surfaces of the cell. In region II (pH 10.6), light was uniformly transmitted through cross polars, reflecting the presence of a twist distortion of $\sim 90^\circ$ within 5CB in this region of the cell. The optical texture of the same cell viewed under parallel polars is shown in Figure 7B and confirms the interpretation of the optical texture shown in Figure 7A. In short, transformation of the acid to salt in region II caused the formation of a twist distortion in this region of the optical cell, whereas no distortion of 5CB was measured in region I (Figure 7C). The boundary between the carboxylic acid and carboxylate regions was easily resolved by the difference in orientations of 5CB with a resolution better than $5 \mu\text{m}$ (Figure 7D and E).

Because LCs are known to contain significant amounts of water,⁴⁶ we considered the possibility that water within LCs may perturb the states of the surfaces presenting acid and salts and thus the orientations of supported LCs. By using Karl Fischer titration, we measured the concentration of water in samples of 5CB and MBBA to be 42 ± 7 and 24 ± 3 mM, respectively.²⁸ To investigate a possible role for water in our experimental observations, we patterned a surface with acid and carboxylate regions (as described above) and then imaged the boundary between the two regions as a function of time. An exchange of water between the surfaces and the bulk of the LC would, we predicted, lead to time-dependent changes in the orientation of the LC in the vicinity of the boundary between the acid and salt groups. We observed, however, no change in the anchoring of the LC in the vicinity of the boundary over a period of 48 h.

4. Discussion

The results reported in this paper demonstrate that LCs can be used to amplify and optically detect the conversion of carboxylic acid groups into sodium carboxylate salts on surfaces. Conversion of the carboxylic acid to its sodium salt is amplified into a change in orientation of 10^4 – 10^5 mesogens per surface-confined acid group, assuming a LC layer thickness of $15 \mu\text{m}$ and one surface-confined acid group in each 20 \AA^2 of the surface. The principal significance of this work is that it establishes the use of LCs for the micrometer-scale imaging of immobilized reactants and products of reversible reactions that are performed by contact of the immobilized species with a solvent phase containing reactants. These results, when combined with past studies describing the use of LCs to image the specific binding to macromolecular species to surfaces, dem-

(46) Kimura, N.; Umemura, J.; Hayashi, S.; Takenaka, T. *J. Mol. Struct.* **1984**, *116*, 153–164.

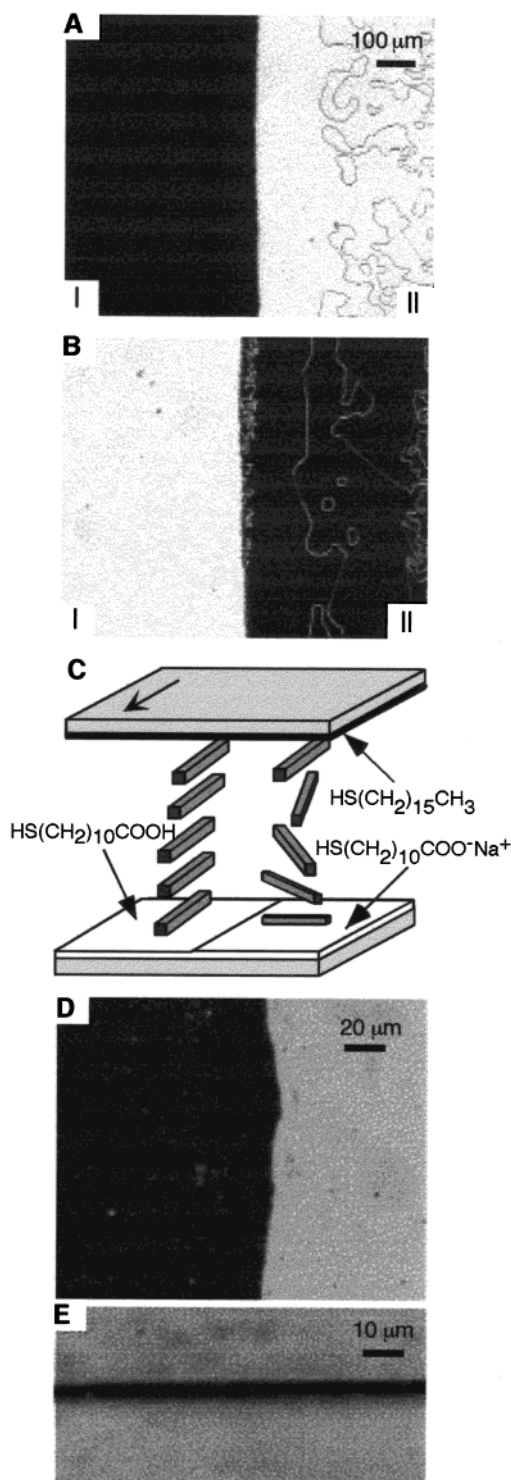


Figure 7. Optical images formed by 5CB supported on surfaces patterned with SAMs presenting carboxylic acid and sodium carboxylate groups. (A and B) Optical textures of 5CB within cells formed with one surface supporting a SAM formed from $\text{H}_3\text{C}(\text{CH}_2)_5\text{SH}$ and an opposing surface supporting a SAM that presents regions of carboxylic acid groups (region I) or sodium carboxylate groups (region II): (A) cross polars and (B) parallel polars. (C) Schematic illustration of the orientations of the LC interpreted from the optical images shown in A and B. (D) High-resolution image of the optical texture of a nematic phase of 5CB at the boundary between regions presenting carboxylic acid and sodium carboxylate groups. (E) High-resolution image of the optical texture of a nematic phase of 5CB supported on a patterned, mixed SAM formed from 45% $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ and 55% $\text{CH}_3(\text{CH}_2)_{11}\text{SH}$. The mixed SAM was patterned by pulling a microdroplet of aqueous solution (pH 12) across the SAM (horizontally). When observed between parallel polars, the 5CB supported on the regions of the mixed SAM contacted with the microdroplet appear dark.

onstrate the general utility of LC-based methods for the study of the interactions between molecular and macromolecular species on surfaces. Whereas the specific binding of antibodies to antigens supported on obliquely deposited gold films was shown to erase the anisotropic roughness of these gold substrates and thereby create nonuniform azimuthal orientations of LCs,⁹ the acid–base reactions reported in this paper cause changes to the structure of surfaces on spatial scales that are small as compared to the size of the nanometer-scale anisotropic roughness of the gold film.¹⁰ The LC orientations reported in this paper are, therefore, uniform ones that are defined with respect to the symmetry of the surface: transformation of immobilized carboxylic acid groups to sodium carboxylate salts triggers a change in the preferred azimuthal orientations of LCs from parallel to perpendicular to the direction of deposition of the gold.

Our past studies have established the presence of nanometer-scale, anisotropic roughness within obliquely deposited gold films such that the elastic energies of supported LCs are minimized by azimuthal orientations of the LCs that are perpendicular to the direction of deposition of the gold.^{10,47} That is, on bare gold films, LCs orient in a direction that is perpendicular to the direction of deposition of the gold. The azimuthal orientations assumed by LCs supported on SAMs presenting sodium carboxylate groups (perpendicular to direction of deposition of gold) do, therefore, likely reflect minimization of elastic distortions within the LCs caused by the anisotropic roughness of the gold films. In contrast, however, on SAMs formed from $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$, both 5CB and MBBA assume an azimuthal orientation that is parallel to the direction of gold deposition. Because this orientation of the LC corresponds to one that is parallel to the direction of maximum roughness, it cannot be understood in terms of minimization of the elastic energy of the LC. Indeed, we have observed the azimuthal orientations of LCs on SAMs supported on obliquely deposited gold films to be specific to both the type and orientation of the functional group presented at the surface of the SAM. For example, whereas the orientations of 5CB on SAMs formed from $\text{Br}(\text{CH}_2)_{11}\text{SH}$, $\text{HO}(\text{CH}_2)_{11}\text{SH}$, or $\text{HOOC}(\text{CH}_2)_{10}\text{SH}$ result in an azimuthal orientation that is parallel to the deposition direction of the gold, on SAMs formed from $\text{NC}(\text{CH}_2)_{11}\text{SH}$ and $\text{HO}(\text{OCH}_2\text{CH}_2)_2(\text{CH}_2)_{10}\text{SH}$, the azimuthal alignment of 5CB is perpendicular to the direction of deposition of the gold.⁴⁸ The influence of the orientation of surface-confined functional groups on the azimuthal orientations of LCs can be illustrated by the behavior of LCs supported on SAMs formed from alkanethiols with odd and even numbers of carbons:⁴⁴ SAMs formed with an odd number of methylene groups orient LCs parallel to the direction of deposition of the gold film whereas SAMs formed with an even number of methylene groups orient LCs perpendicular to the direction of deposition of the gold. While the origin of the specificity of the LC to the type and orientation of the ω -functional group is the subject of an ongoing investigation based on second harmonic generation and sum-frequency spectroscopy, we note that conversion of a carboxylic acid to a sodium carboxylate salt will eliminate possible hydrogen-bonding interactions between the hydrogen of the carboxylic acid and the nitrogen of the nitrile group of 5CB.⁴⁹ We speculate that loss of hydrogen-bonding interactions between 5CB and the COOH-terminated SAMs upon conversion of the acid to the salt may underlie the discontinuous change in the azimuthal orientations of LCs that we report in this paper.⁵⁰

(47) Gupta, V. K.; Abbott, N. L. *Langmuir* **1996**, *12*, 2587–2593.

(48) Shah, R. R.; Poquechoque, R.; Abbott, N. L. Unpublished results.

We do not know the geometry of the hydrogen-bonded complex formed between 5CB and the carboxylic acid groups at the surface of the SAM.⁵¹

A significant result of the study reported herein is the success of simple and general procedures based on LCs that permit determination of the states of molecules on surfaces that undergo fast and reversible reactions upon contact with solvent phases. Whereas LCs have been used in past studies to detect specific binding of proteins to surfaces in which the binding was strong and essentially irreversible,⁹ conversion of a carboxylic acid to its sodium carboxylate salt by immersion in a buffered aqueous solution is a rapidly reversed reaction. The experimental procedures reported in this paper involve the displacement of buffered aqueous solutions from the surfaces of SAMs by using a stream of nitrogen: the surfaces were not rinsed with water or other solvents prior to contact with the LC. The success of this simple experimental procedure contrasts with past studies involving similar protocols and polymeric surfaces presenting carboxylic acid groups.²¹ Past studies using high- or low-density polyethylene (HDPE or LDPE) functionalized with carboxylic acid groups and procedures similar to those used by us have reportedly substantial difficulty controlling the extent of conversion of carboxylic acid groups to their corresponding salts.²¹ The extent of conversion of acid to salt was found to depend on a number of factors, including the time of immersion in the aqueous solutions. We believe several factors contribute to the difficulties reported in these past studies. First, the polymer–solution interface is known to be a mobile and diffuse one that extends over micrometers. Second, polymeric interfaces can reconstruct over time, especially when placed into contact with solvents such as water. Third, in addition to the carboxylic acid groups, these polymers presented alcohols, aldehydes, ketones, and esters at their surfaces. In contrast, when using SAMs formed from HOOC(CH₂)₁₀SH, we observed reproducible and stable conversion of the carboxylic acid to its salt. Our results based on ellipsometry, XPS, and FTIR demonstrate that this procedure (i) leads to an extent of conversion of acid to salt that can be controlled by the pH of the aqueous solution

(49) Hydrogen bonding between the nitrile group of 5CB and the COOH group of *p*-ethylbenzoic acid and water has been observed by using IR spectroscopy by Asaba et al.⁵³ Kimura et al.⁴⁶ observed hydrogen bonding between the nitrile group of 4-cyano-4'-octylbiphenyl (8CB) and water using polarized IR spectroscopy. The nitrogen in MBBA may also be involved in hydrogen-bonding interactions with carboxylic acid groups of SAMs.

(50) Discontinuous changes in the azimuthal orientations of LC on surfaces are well known and can be caused by an azimuthal potential energy function that possesses two energetic minima. As the relative magnitudes of these minima change, the orientation of the liquid crystal shifts discontinuously from the angle corresponding to one minimum to the angle corresponding to the other minimum. (Jérôme, B. *Rep. Prog. Phys.* **1991**, *54*, 391–451.) Our experimental results indicate the likely presence of two energetic minima—one minimum is controlled by the structure of the gold film induced by the oblique deposition (e.g., the minimum associated with elastic interactions with the statistically corrugated surface),¹⁰ and the other minimum is controlled by short-range interactions (e.g., hydrogen bonding) between the SAM and 5CB.⁵¹ Conversion of COOH to COONa is accompanied by the loss of the short-range interactions between the 5CB and COOH and thus results in an orientation of 5CB that is dictated by the structure of the gold.

(51) Three observations lead us to believe that hydrogen bonding between 5CB and SAMs formed from HOOC(CH₂)₁₀SH influences the orientation assumed by 5CB on these surfaces. First, on bare gold films (in absence of a hydrogen-bonding interaction), 5CB orients in a direction perpendicular to the direction of deposition of the gold film. Second, on SAMs formed from HOOC(CH₂)₁₀SH, where hydrogen-bonding interactions between 5CB and the terminal carboxylic acid group of the SAM can influence the orientation of 5CB, we observe 5CB to orient parallel to the direction of deposition of the film of gold underlying the SAM. Third, conversion of the terminal carboxylic acid groups to sodium carboxylate, which eliminates the possibility of hydrogen-bonding interactions between the SAM and 5CB, results in an orientation of 5CB that is perpendicular to the direction of deposition of the gold film.

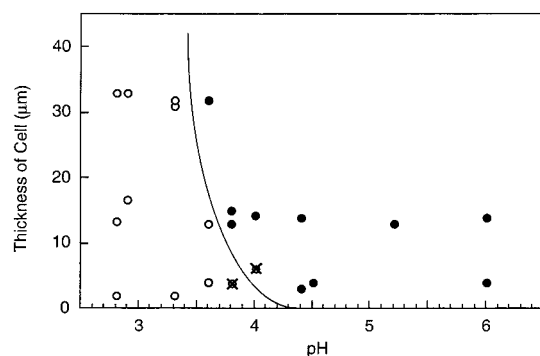


Figure 8. pH of aqueous solutions that triggered twist distortions within a nematic phase of 5CB as a function of the thickness of the optical cell. Optical textures of 5CB were observed between cross polars in cells with one surface supporting a SAM formed from H₃C(CH₂)₁₅SH and an opposing surface formed from HOOC(CH₂)₁₀SH that was pretreated by immersion in aqueous solutions buffered between pH 2.6 and 6. Open data points indicate the absence of twist within the LC cell; filled points indicate formation of twisted distortions within the LC; crosses indicate cells that possessed localized regions of twist. The solid line indicates the approximate location of the boundary between untwisted and twisted states of the LC.

used to pretreat the SAMs and (ii) does not lead to the deposition of excess salt crystals on these surfaces over a wide and useful range of pH's.

Although several past studies have characterized the electrical double layer and resulting electrostatic forces in aqueous solutions near SAMs presenting carboxylic acid groups, few groups have characterized these surfaces following their removal from buffered aqueous solutions.³⁰ Following the removal of a SAM formed from HOOC(CH₂)₁₀SH from an aqueous solution buffered at pH 8.5, our XPS results indicate that $\sim 50 \pm 8\%$ of the immobilized carboxylic acid groups are converted to their salts. For comparison, for SAMs immersed in aqueous solutions at pH 7.7, Hu and Bard^{32a} have interpreted force–distance curves to indicate that 50% of the acid groups on SAMs formed from HOOC(CH₂)₃SH are ionized. The close agreement between the extent of ionization of the acid groups in solution and extent of conversion of acid to salt following removal from solution leads us to conclude that the process of removal of the SAMs from the aqueous solutions does not lead to gross changes in the extent of conversion of acid to salt.

Because the 5CB used in our experiments contained a concentration of water of 42 ± 7 mM, we considered it possible that the water within 5CB would cause immobilized carboxylic acid groups to ionize or sodium carboxylate groups to protonate. Two experimental results suggest, however, that water within the LC does not significantly perturb the states of SAMs used in our experiments. First, we examined the orientations of LCs at the boundary regions of surfaces patterned with acid and carboxylate groups (as described above). If exchange occurred between the water in the LC and the SAMs, we would expect the compositions of the patterned regions of the surface to equilibrate with each other and the LC to adopt a uniform orientation at the boundary of the patterns. We observed the edges of the patterns formed by LCs to be sharp and stable for days. Second, we varied the thickness of the LC layer between 2 and 30 μm and measured the threshold pH that caused a change in orientation of the LC. If exchange of water between the LC and the SAM caused a change in the state of the SAM, we would expect the threshold pH to change significantly with the thickness of the cell. Instead, the threshold pH shifted by only 0.5 pH unit with the thickness of the cell (Figure 8). We offer two plausible reasons as to why water within the LC does

not measurably perturb the states of these surfaces and the orientations of supported LCs. First, when using a LC cell with a thickness of 20 μm , we calculated there to be approximately 5000 water molecules within the LC per COOH group on the surface (a ratio of water to acid that is equivalent to a bulk aqueous solution containing 10 mM acid). The buffering effect of the COOH/COO⁻Na⁺ is large at these ratios of acid to water.⁵² Second, past studies⁴⁶ based on IR spectroscopy suggest that water molecules are hydrogen bonded to the nitrile groups of smectic A phases of 4-cyano-4'-octylbiphenyl (8CB). These measurements were performed using a LC with roughly 1 water molecule for each 90 molecules of 8CB, which is similar to the water content of the 5CB used in our experiments (approximately 1 water molecule for each 100 molecules of 5CB). These results suggest that water within LC phases of 5CB may be complexed with the LC and thus possess a low activity.

Finally, we mention that the principal method we propose for transducing the orientations of LCs into optical signals is based on formation of twist distortions within the LC. Because the energy stored within the twist distortions depends inversely on the thickness of the optical cell,^{6a} we determined if the thickness of the optical cell influenced the threshold conversion of acid to salt required to trigger formation of the twist deformation. As mentioned above, we observed the threshold pH to differ by 0.5 pH unit when the thickness of an optical cell was increased from 2 to 30 μm (Figure 8). The conclusions

(52) Assuming ideal solution behavior, we calculated the extent of ionization of a carboxylic acid when added to an aqueous solution at pH 7 such that the ratio of water to acid is 5000. Assuming a $pK_{1/2}$ of 7.7 for the acid (as reported by Bard and co-workers for a surface-confined carboxylic acid^{32a}), the extent of ionization of the acid is calculated to be 0.14%. We have also calculated the extent of protonation of a sodium carboxylate salt when added to an aqueous solution at a ratio of water to salt of 5000. In this case, the extent of protonation of the carboxylate ion was 0.7%. Similarly, addition of an equal molar ratio of acid and carboxylate salt to water results in a solution containing 50.001% acid and 49.999% salt.

(53) Asaba, K.; Igarashi, A.; Kobinata, S. *Jpn. J. Appl. Phys.* **1998**, *37*, 6482–6485.

of this simple experiment are two-fold. First, manipulation of the thickness of the cell is unlikely to form the basis of a useful method to change the extent of a reaction which causes a change in orientation of a LC. Second, when using optical cells and mixed SAMs to determine the extent of conversion of acid to salt, a high level of control over the thickness of the cell is not necessary.

5. Conclusions

The principal conclusion of the experiments reported in this paper is that LCs can be used to image (with micrometer resolution) reactants and products of reversible chemical reactions on surfaces that are performed by contact of surface-confined species with reactants dissolved in solution. The model reaction used in this investigation—conversion of immobilized carboxylic acid groups to sodium carboxylate salts—demonstrates the usefulness of simple and general LC-based procedures for mapping interactions between small molecular species on surfaces. The results reported herein also provide an approach to LC-based methods that permit quantitation of the extent of conversion of reactants to products on surfaces. For example, we report the design of surfaces that trigger twist distortions within LCs when the extent of an acid–base reaction reaches specified values between 8% and ~100%.

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