Macromolecules **2007,** *40,* 6967−6972

Hydrogen-Bonded Multilayers of Poly(carboxybetaine)s

Eugenia Kharlampieva, Denis Pristinski, and Svetlana A. Sukhishvili*

Department of Chemistry and Chemical Biology, Stevens Institute of Technology, Hoboken, New Jersey 07030

Received May 22, 2007; Revised Manuscript Received July 12, 2007

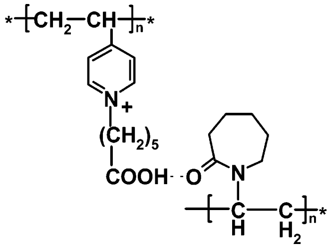
ABSTRACT: Using a layer-by-layer (LbL) sequential adsorption technique, we studied the construction of hydrogen-bonded multilayers of a weak polycarboxybetaine (poly-*N*-*ω*-carboxypentyl-4-vinylpyridinium bromide (PCB5)) with poly(*N*-vinylcaprolactam) (PVCL). Self-assembly occurred at low pH values where carboxylic groups of PCB5 units were protonated and capable of binding with hydrogen-accepting groups of PVCL. Multilayers could not be constructed, however, in 0.01 M buffer solutions at pH 2.5, and potassium halide salts were required to promote multilayer deposition. Concentrations of halide salts at which deposition of PCB5/PVCL multilayers occurred decreased in the series KCl > KBr > KI, following the Hofmeister series for binding of anions with PCB5 chains. Using a *N*-vinylcaprolactam copolymer which contained amino groups (PVCL-NH_2), PCB5/PVCL-NH_2 multilayers were constructed and covalently cross-linked, producing surface-attached hydrogels. The hydrogels showed typical polyelectrolyte salt-induced contraction at pH 2.5, where PCB5 units were cationic, but "antipolyelectrolyte" behavior, i.e., swelling in salt solutions at pH 7.5, where PCB5 units were zwitterionic. PCB5/PVCL-NH_2 surface hydrogels were resistant to adsorption of negatively charged proteins and heparin at pH 7.4 in 0.15 M NaCl solutions. The produced PCB5/PVCL films and cross-linked PCB5/PVCL-NH_2 surface hydrogels might find applications as erasable films or as zwitterion-containing bioinert coatings, respectively.

Introduction

Nature shows many examples of self-assembly of highly functional structures such as membranes, proteins, or nucleic acids using noncovalent interactions. In DNA, the negatively charged sugar−phosphate backbones are bound via hydrogen bonding between the base pairs, which is additionally stabilized by hydrophobic or stacking interactions. Using approaches learned from nature, cooperative self-assembly through combined hydrogen-bonding, stacking, and hydrophobic interactions has been recently used to produce non-covalently-linked synthetic helical structures,[1] nanotubes,[2] and supramolecular membranes[3] using non-polymeric building blocks. Synthetic polymers have also been self-assembled by means of hydrophobic interactions and hydrogen bonding in aqueous solutions to produce conducting polymer aggregates in solution[4] or at surfaces.[5] In the latter case, sequential alternating adsorption of polymers from solution (or layer-by-layer (LbL) technique) to produce ultrathin films has recently received much attention. Hydrogen-bonded LbL films were first reported by Stockton and Rubner[5] and Wang et al.[6] (see recent feature article for details[7]). We have earlier demonstrated that water-soluble uncharged molecules capable of intermolecular hydrogen-bonding interactions can be self-assembled at surfaces using the LBL approach and then controllably dissolved at a higher pH value.[8,9] Routes to stabilize hydrogen-bonded self-assembly via thermal, photochemical,[10] or chemical cross-linking[11−13] as well as the ability to deposit and stabilize hydrogen-bonded polymer layers on spherical substrates[11−16] have been recently reported by several groups. Stabilized hydrogen-bonded capsules are being explored for encapsulating various chemicals and controlled release of encapsulated cargo.[13,14]

Hydrogen bonding has also been used to promote LbL growth of like-charged polyelectrolytes. Recently, Caruso's group reported LbL deposition of negatively charged polynucleotides at surfaces based on DNA hybridization driven by hydrogen bonding.[17] Note also that earlier attempts to assemble positively charged polyelectrolytes resulted in very limited film growth if hydrogen bonding was not involved in the assembly.[18] Very recently, Schlenoff and co-workers studied LbL deposition of similarly charged hydrogen-bonding homopolynucleotides (poly(adenylic acid) and poly(uridylic acid)) and found that these two molecules failed to produce films, unless they were coassembled into films with a polycation.[19]

Scheme 1. Hydrogen Bonding between PCB5 and PVCL Units



In this work, we explore hydrogen-bonded LbL self-assembly of a weak zwitterionic polybetaine (PB) with a neutral polymer in aqueous environment. Interest to incorporate PBs into thin layers is greatly due to PBs' improved biocompatibility,[20] antibacterial properties,[21] and their utility for building bioinert coatings.[22,23] The unique properties of PBs are often explained by the zwitterionic nature of the PB head group which mimics naturally existing phosphorylcholine groups commonly found in cell membranes. We have recently studied electrostatic self-assembly of a weak polycarboxybetaine (poly-*N*-*ω*-carboxypentyl-4-vinylpyridinium bromide (PCB5)) within LbL films with polyanions and/or polycations at slightly acidic and neutral pH values.[24] Here, we focus on hydrogen-bonding self-assembly of a PCB5 at low pH values where carboxylic groups of PCB5 units are protonated and are able to interact with hydrogen-accepting units of a neutral polymer, such as those of poly(*N*-vinylcaprolactam) (PVCL) shown in Scheme 1. Because of the

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* To whom correspondence should be addressed.

10.1021/ma071152i CCC: \$37.00

cationic charge of PCB5 chains in these conditions, deposition of PCB5/PVCL multilayers did not occur at low concentrations of small ions, unless poly(styrenesulfonate) (PSS) was introduced as a third component in self-assembly, and increased concentrations of salts were required to promote successful multilayer growth. We also found that, similar to hydrogen-bonded films of poly(carboxylic acid)s and a neutral polymer, the produced films can be erased from a solid surface by exposure to solutions with higher pH values due to deprotonation of carboxylic groups of PCB5 units. However, using an amino-group-containing *N*-vinylcaprolactam copolymer, hydrogen-bonded PCB5/PVCL films can be covalently cross-linked, producing surface-attached hydrogels which did not desorb at neutral and basic pH values. Here, we explore the effect of salt on swelling of PCB5-containing surface hydrogels at low or neutral/basic pH values, where PCB5 units are cationic or zwitterionic, respectively, and show that the salt has opposite effect on hydrogel swelling in these two different pH ranges.

Experimental Section

Materials. PVCL (MW 1800) and PSS (MW 500 000) were purchased from Polymer Source and Scientific Polymer Products, Inc., respectively. PCB5 with five methylene groups separating N^+ and COO^- was synthesized by reaction of poly(4-vinylpyridine) (P4VP, Aldrich, MW 200 000) with ω-bromocarboxylic acid (Aldrich) as described elsewhere.[25] Halide salts (KCl, KBr, and KI) were obtained from Alfa Aesar, Co. Millipore (Milli-Q system) filtered water with a resistivity 18.2 MΩ was used in all experiments. D_2O with 99.9% isotope content was purchased from Cambridge Isotope Laboratories and was used as received. For $pH > 4.5$, 0.01 M K_2HPO_4 and KH_2PO_4 buffer solutions (Fisher Scientific) were used. For lower pH values, 0.01 M KH_2PO_4 solution was adjusted with HCl.

Synthesis of PVCL-NH$_2$. Copolymer of *N*-vinylcaprolactam (VCL) and glycidyl methacrylate (GMA) was synthesized by free-radical solution polymerization in dioxane using AIBN as initiator as described in our recent publication.[26] The produced copolymer, poly(VCL-*co*-GMA) (10% GMA, MW 19 000), was reacted with 100-fold excess of ethylenediamine (with respect to molar GMA content in the copolymer) in water at room temperature for 24 h to produce an amino-containing copolymer PVCL-NH$_2$. Excess ethylenediamine was removed by 72 h dialysis, followed by freeze-drying of the copolymer.

Deposition of PSS/PCB5/PVCL and PCB5/PVCL Multilayers. Multilayers were prepared on surfaces of silicon wafers or onto a Ge or Si attenuated total reflection (ATR) crystal by sequential deposition from polymer solutions at pH 2.5. The ATR crystals were precleaned through 2 h exposure to UV radiation, rinsed with deionized water, copiously rinsed with Milli-Q water, dried in nitrogen stream, and finally oxygen plasma-treated for 2 min. For ellipsometry studies, deposition of multilayers was performed from aqueous polymer solutions onto flat silicon wafers using a home-built robotic system. In a typical experiment, the substrates were alternately exposed to 0.1 mg/mL polymer-buffered solutions at pH 2.5 for 10 min followed by two 30 s intermediate rinsing steps with water at pH 2.5. Before the measurements, films were blow-dried with ultrapure nitrogen. For in situ ATR-FTIR (Fourier transform infrared spectroscopy in ATR mode) studies, multilayers were prepared using a procedure similar to that described above, except that multilayers were constructed on the surface of an oxidized Ge or Si ATR crystal installed within a flow-through ATR-FTIR stainless steel cell. The deposition was performed in situ using D_2O polymer solutions.

Deposition of PCB5/PVCL and PSS/PCB5/PVCL films was performed at surfaces of oxidized Si or Ge crystals, respectively. In both cases, PCB5 was allowed to adsorb at the crystal surface at pH 6. After that, the pH was changed to 2.5, and deposition was continued at this lower pH value starting from PVCL or PSS for PCB5/PVCL or PSS/PCB5/PVCL films, respectively.

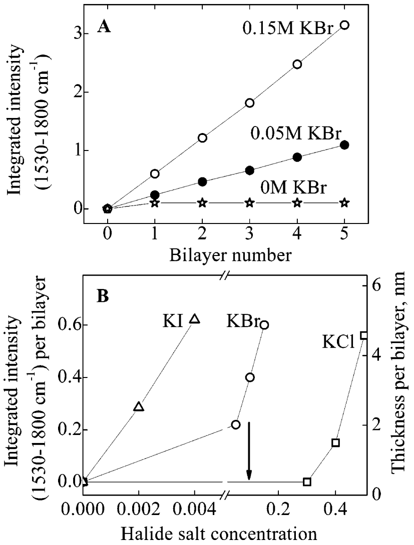

Figure 1. (A) Evolution of total integrated ATR-FTIR intensities in the 1530−1800 cm^{-1} range (see Experimental Section) during in situ monitoring of deposition of (PCB5/PVCL)$_5$ films from 0.15 M KBr (open circles) and 0.05 M KBr (filled circles) and 0 M KBr (open stars) solutions at pH 2.5. (B**)** Integrated 1530−1800 cm^{-1} ATR-FTIR intensities measured in situ (left axis) and ellipsometric thicknesses of dry films (right axis) per PCB5/PVCL bilayer within (PCB5/PVCL)$_5$ films deposited from KI (triangles), KBr (circles), and KCl (squares) solutions at pH 2.5.

ATR-FTIR Measurements. In situ ATR-FTIR experiments were done with a Bruker Equinox-55 Fourier transform infrared spectrometer equipped with a narrow-band mercury cadmium telluride detector. The ATR Si or Ge crystal was installed within a flow-through stainless steel cell. The ATR crystals were rectangular with dimension of 50 mm × 20 mm × 2 mm (Harrick Scientific) whose beam entrance and exit surfaces were cut at 45°. PCB5/PVCL and PSS/PCB5/PVCL multilayer deposition was inferred from integrated intensities in the region 1530−1800 cm^{-1}, which correspond to stretching vibrational bands of carboxylic groups (centered at 1700 cm^{-1}) and pyridinium ring in PCB5 (centered at 1640 cm^{-1}) as well as to the 1640 cm^{-1} amide stretching band of the caprolactam ring. In the case of PSS/PCB5/PVCL films, Ge crystal instead of Si crystal was used to enable observation of PSS-related vibrational bands at 1126, 1036, and 1009 cm^{-1}. Although ATR-FTIR measurements of thin films allow determining the amounts adsorbed after appropriate calibration of signal intensity using solutions of nonadsorbing polymers,[27] here we did not follow this procedure, and the amounts adsorbed were determined by ellipsometry.

Phase-Modulated Ellipsometry. Ellipsometry measurements were performed using a home-built, single-wavelength, phase-modulated ellipsometer at 65° angle of incidence.[28] Ellipsometry substrates were low-dopant Si wafers (Virginia Semiconductor, Inc.) with naturally grown SiO_2 layer. Optical properties of the substrates and oxide layer thicknesses were determined prior to polymer deposition. During analysis the refractive index of all dry polymer films was fixed at a value of 1.5.

For salt concentrations higher than 0.1 M, ellipsometric measurements of dry film thicknesses could not be performed because of light scattering from the surface, caused by deposition of salt after drying of solutions. However, in the case of KI, when PCB5/PVCL films could be deposited from low KI concentrations (1−10 mM), ellipsometric measurements of dry films were obtained and correlated to integral absobances in the 1530−1800 cm^{-1} range obtained by ATR-FTIR. Specifically, the integrated ATR-FTIR intensity value of 0.6 in this region corresponded to ∼5 nm of dry film thickness (assuming refractive index of 1.5). The correlation was used to reconstruct ellipsometric thicknesses for films deposited from KBr and KCl solutions (shown in Figure 1).

Measurements of swelling of PCB5/PVCL-NH$_2$ hydrogels were performed in situ using a custom-made cylindrical flow-through glass cell. To obtain the swollen film thickness, the cell was filled with 10 mM potassium phosphate buffer at different pH values and ionic strength, and measurements were taken after the hydrogel was allowed to equilibrate for 3 min. The procedure for determination of the swollen film thickness and refractive indexes was described in our earlier publication.[28] To check the consistency of the in situ ellipsometry data interpretation, measured values of refractive index of a swollen film n_F were used to calculate the polymer content η using the equation[29] $\eta = (n_F - n_S)/(\rho\ dn/dc)$, where d*n*/d*c* is the average refractive index increment for the surface hydrogel film, ρ is the density of dry polymer film, taken as 1 g/mL, and n_S is solution refractive index depending on the potassium halide concentration.[30] The d*n*/d*c* for the swollen PCB5/PVCL-NH_2 multilayers was estimated to be 0.20 mL/g using reported values for refractive index increments of aqueous solutions of polymer film components, i.e., d*n*/d*c* of 0.21,[31] 0.171,[32] and 0.21 mL/g[33] for PVCL, PAA, and quaternized P4VP solutions, respectively (all values reported for 25 °C). Then dry film thickness was compared with the product of swollen film thickness and swollen film polymer content. For all measurements performed at pH below 6 and all potassium halide concentrations, the results were in a good agreement, with a discrepancy less than 10%.

Cross-Linking of PCB5/PVCL-NH_2 Films. To enhance the surface adhesion of PCB5/PVCL-NH_2 films to the substrate, a precursor layer of two bilayers of PAH/PMAA was deposited and stabilized by thermal cross-linking in an oven at 130 °C for 1 h.[34] (PCB5/PVCL-NH_2)$_5$ films were then deposited from 0.2 M KBr solution at pH 2.5. Cross-linking was performed via standard carbodiimide chemistry by adding 1-ethyl-3-(3-(dimethylamino)-propyl)carbodiimide hydrochloride (EDC) and *N*-hydroxysulfosuccinimide sodium salt (5 mg/mL of each) at pH 4.5. The cross-linking time was 12 h. To remove activation agents, cross-linked multilayers were then exposed to 10 mM phosphate buffer solution at pH 7.5 for 2 h.

Results and Discussion

Figure 1 shows that multilayers of PCB5 can be constructed through alternating adsorption with poly(vinylcaprolactam) (PVCL) at low pH values and that the deposition is largely controlled by the concentration and type of small ions. It is interesting that weakly hydrogen-bonding polymers which contain ether groups, such as poly(ethylene oxide) (PEO) and poly(vinyl methyl ether) (PVME), or secondary amide groups, such as poly(*N*-isopropylacrylamide) (PNIPAM), did not produce multilayers with PCB5 even at high concentrations of salts. On the other hand, self-assembly of PCB5 with poly(vinylpyrrolidone) (PVPON), a homologue of PVCL which lacks two methylene groups in the ring, also failed. These experiments show that both the intrinsic strength of hydrogen-bonding units and additional hydrophobic stabilization of intermolecular hydrogen bonding were critical for multilayer growth. Even with strongly interacting PVCL, PCB5/PVCL multilayers could not be deposited from low salt solutions. Figure 1A shows that PCB5/PVCL films could not be deposited in pure D_2O or water at pH 2.5, but addition of KBr promoted film deposition, and that film growth was linear for various salt concentrations. As directly monitored with in situ ATR-FTIR, carboxylic groups of PCB5 remained completely protonated during deposition at pH 2.5, in agreement with typical pK_a values reported of PCBs.[35−37] For example, McCormick has reported pK_a of 3.6 for carboxylbetaine with three methylene groups between the carboxylic group and the positive charge in the polymer unit.[37] As shown in Figure 1, in a completely cationic state, self-assembly of PCB5 was still possible through hydrogen bonding of carboxylic groups with caprolactam rings when electrostatic repulsions between PCB5 positive charges were screened by increased concentration of KBr.

Figure 1B shows that deposition of PCB5/PVCL films was also strongly controlled by the type of anion. In KCl solution, concentrations larger than 0.3 M (corresponding to Debye screening length smaller than 6 Å) were required to promote PCB5/PVCL film deposition, while film self-assembly occurred at much smaller concentrations of KBr. In KI solutions, films could be deposited at very small concentrations of salt, ∼1−10 mM.

The salt effect shown in Figure 1 reflects stronger condensation of chaotropic, less hydrated anions on the PCB5 chains (as compared to kosmotropic, more hydrated anions) and is similar to reported order of specific binding of anionic counterions with synthetic polycations or synthetic proteins which follows the Hofmeister series.[38,39] An alternative explanation of the effect of salt on solubility of a neutral polymer can be suggested, as the salt effect on the lower critical solution temperature (LCST) of PNIPAM[40,41] and PVCL[42] also follows the anion Hofmeister series,[43,44] with kosmotropic anions decreasing the LCST.[40−42] Specifically, I^- was found to increase the solubility and raise the LCST of PVCL solutions, so as Br^- at concentrations lower than 0.3 mg/mL.[42] However, this effect acted in an opposite direction than the one found for PCB5/PVCL multilayers and was very small (∼1−2 °C) for concentrations of halide salts shown in Figure 1B. Consequently, the above scenario could not explain the strong ionic specificity of multilayer deposition. Instead, specific ionic binding to a polycation agrees well with the data in Figure 1B. In this work, stronger binding of less hydrated and more polarizable anions with PCB5 was also illustrated by the following concentrations of halide salts required for salting-out of PCB5 from 0.1 mg/mL solutions at pH 2.5: 1.6 M KCl, 0.5 M KBr, and 0.03 M KI. Importantly, for all salt types, the onset of film deposition occurred at much lower salt concentrations than those required for salting-out of PCB5 from solution. Strong specificity of anion binding with cationic polyelectrolytes has been found for the case of polyelectrolyte complexes (PEC) in solution, where the capacity of anions to cause PEC dissociation followed the order $I^- > Br^- > Cl^- \gg F^-$.[45−47] Such specificity is likely induced by dispersion forces between the anions and polymer chains. The importance of dispersion forces in the specificity of anion binding has been recently demonstrated in a direct measurement of the degree of disordering of hydrophobic regions of a monolayer caused by different anions.[48]

A specific counterion effect was also observed with electrostatically self-assembled polyelectrolyte multilayers[49,50] where multilayer thickness increased in the order NaF < NaCl < NaBr.[49] The order reflects stronger condensation of less hydrated anions on the polycation chains, leading to larger amounts of polymers deposited within the film through a charge-compensation mechanism. Although the order of the anion effect on multilayer deposition found in this work is similar to that observed for electrostatically associated PEMs, the counterion effect is much stronger in the case of hydrogen-bonded multilayers. The arrow in Figure 1B illustrates that a change of anion type from Cl^- to Br^- at a constant concentration of ions (e.g., 0.15 M) results in an "on−off" transition in multilayer growth. Such strong sensitivity of deposition of hydrogen-bonded multlilayers to the counterion type and concentration results from the fact that binding between PCB5 and PVCL chains occurred through hydrogen bonding, rather than ionic pairing, and electrostatic repulsions between cationic PCB5 units which opposed multilayer buildup.

Another way to promote PCB5/PVCL binding at surfaces at low pH is to co-self-assemble this polymer pair with a

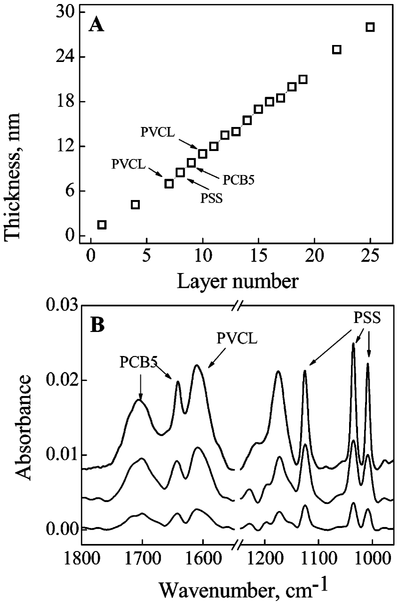

Figure 2. Sequential deposition of hybrid $(PSS/PCB5/PVCL)_n$ films at pH 2.5 as measured by ellipsometry with dry films at the surfaces of silicon wafers (A) or by in situ ATR-FTIR during film deposition from D_2O solutions on a Ge ATR crystal (B, $n = 1$, 3, and 5 from bottom to top, respectively). Arrows in panel B point to vibrational peaks associated with protonated carboxylic groups (−COOH) and pyridinium rings (N^+) of PCB5 centered at 1700 and 1640 cm^{-1}, respectively; an absorption band associated with stretch vibrations of C=O in PVCL at 1610 cm^{-1}; and PSS vibrational peaks at 1125, 1035, and 1008 cm^{-1}.

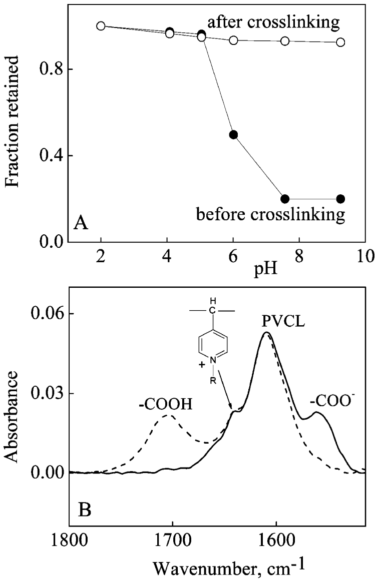

Figure 3. (A) pH stability of $(PCB5/PVCL\text{-}NH_2)_5$ films before (filled circles) and after cross-linking (open circles) measured with in situ ATR-FTIR. The stability of films before and after cross-linking was performed in 0.2 M KBr buffered D_2O solutions and buffered D_2O solutions without KBr, respectively. (B) Representative ATR-FTIR spectra of a cross-linked $(PCB5/PVCL\text{-}NH_2)_5$ film at pH 2.5 (dashed line) and pH 7.5 (solid line). Vibrational peaks associated with protonated carboxylic groups (−COOH), ionized carboxylic groups ($-COO^-$) of PCB5, and with PVCL are centered at 1700, 1550, and 1610 cm^{-1}, respectively. The arrow points to a vibrational peak of PCB5 pyridinium rings centered at 1640 cm^{-1}.

polyanion, PSS, which compensates positive charge within the film. A similar strategy of compensating extra charge of hydrogen-bonding polymers by introducing oppositely charge polycation was recently employed[19] to introduce similarly charged homopolynucleotides into thin films. Figure 2A shows that ellipsometric thickness of dry three-component $(PSS/PCB5/PVCL)_5$ films linearly increased with layer number when a hybrid film was constructed at pH 2.5 and zero salt concentration. Figure 2B illustrates evolution of ATR-FTIR spectra in PSS/PCB5/PVCL system during deposition of such film in situ in D_2O solutions at pH 2.5. Integrated intensities in 1530−1800 cm^{-1} (PCB5 and PVCL) as well as in 1050−990 cm^{-1} (PSS) regions also showed a linear increase with the number of trilayers. The successful deposition of PCB5 and PVCL at surfaces in the presence of PSS is caused by charge compensation between PCB5 and PSS molecules. Deposition of PCB5 on PSS-topped film results in neutralization of positively charged units of PCB5 with PSS negative charge and enables protonated carboxylic groups of PCB5 to form hydrogen bonds with PVCL. Further adsorption of PSS chains on PVCL-topped film then occurs due to interaction with the underlying PCB5 layer.

Similar to other hydrogen-bonded films studied earlier,[51] hydrogen bonds between PCB5 and PVCL dissociate at high pH values due to ionization of PCB5 carboxylic groups. To stabilize PCB5/PVCL multilayers at high pH values, covalent cross-links were introduced within the film. For that purpose, a $PVCL\text{-}NH_2$ copolymer, which contained ~10% of units with primary amino groups available for cross-linking with carboxylic groups of PCB5, was employed. (See Experimental Section for $PVCL\text{-}NH_2$ synthesis and cross-linking procedures.) Deposition of $PCB5/PVCL\text{-}NH_2$ films in 0.2 KBr solutions at pH 2.5 showed linear growth, with bilayer thickness slightly smaller than that in PCB5/PVCL films deposited in the same conditions (3.5−4 nm vs 6 nm, respectively). The difference is due to the positive charge resulting from protonated amino groups in the $PVCL\text{-}NH_2$ chains, which additionally opposes hydrogen-bonded self-assembly. Figure 3A contrasts relative ellipsometric thicknesses of dry $(PCB5/PVCL\text{-}NH_2)_5$ non-cross-linked and cross-linked films. While non-cross-linked $(PCB5/PVCL\text{-}NH_2)_5$ films deposited at pH 2.5 in 0.2 M KBr solutions dissolved when exposed to 0.2 KBr solutions with higher pH values, cross-linked $(PCB5/PVCL\text{-}NH_2)_5$ films showed negligible mass loss in solutions with high pH values (Figure 3A). The result was also confirmed by the in situ ATR-FTIR technique, which showed no mass loss for cross-linked $(PCB5/PVCL\text{-}NH_2)_5$ films exposed to pH 7.5 (Figure 3B). In addition, cross-linked films retained a large amount of carboxylic groups which showed reversible protonation (the peak centered at 1550 cm^{-1}) and deprotonation (the peak centered at 1700 cm^{-1}) when exposed to lower and higher pH values. A broad band associated with amide I vibrations and usually observed in the 1620−1660 cm^{-1} region is not clearly seen in the spectrum of cross-linked films due to an overlap with bands of PCB5 and PVCL centered at 1640 and 1610 cm^{-1}, respectively.

Swelling of cross-linked $PCB5/PVCL\text{-}NH_2$ films in salt solutions with various pH values were then explored using in situ phase-modulated ellipsometry. The top panel in Figure 4 shows that, when exposed to low-salt solutions, the cross-linked $(PCB5/PVCL\text{-}NH_2)_{10}$ film swelled from its dry thickness of 39.5 nm by a factor of ~2.5 at pH 2.5 and by a factor of ~3.5 at pH 7. Similar swelling profiles with swelling ratios were obtained for $(PCB5/PVCL\text{-}NH_2)_5$ films with a dry thickness of 23 nm. The bottom panel in Figure 4 shows that measured values for the film refractive index correlate with the degree of film swelling. The smaller degree of swelling of $PCB5/PVCL\text{-}NH_2$ hydrogels at low pH values compared to high pH values is probably due to partial association of polymer units within the hydrogel, i.e., formation of hydrogen-bonded dimers of protonated carboxylic groups of PCB5 units. The overall high degree of swelling in low-salt solutions is consistent with the absence

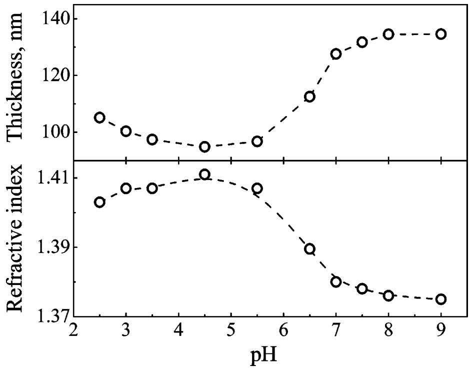

Figure 4. In situ ellipsometry measurements of thickness (top panel) and refractive index (bottom panel) of swollen (PCB5/PVCL-NH_2)$_{10}$ surface hydrogels at various pH values. Dry film thickness was 39.5 nm. 10 mM potassium phosphate buffer was used for pH > 4.5, and for lower pH values, 10 mM KH_2PO_4 was adjusted with HCl.

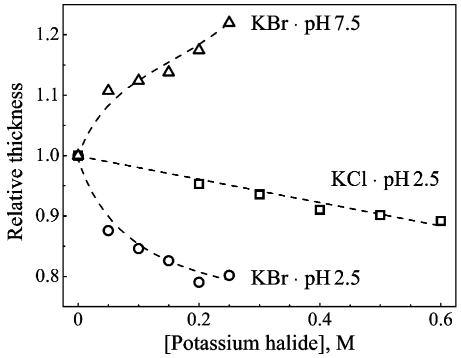

Figure 5. Effect of potassium halide salts on swelling of (PCB5/PVCL-NH_2)$_5$ surface hydrogel films determined by in situ ellipsometry: KBr at pH 2.5 (circles), KCl at pH 2.5 (squares), and KBr at pH 7.5 (triangles). Dashed lines are guide for the eye only. 10 mM potassium phosphate buffer was used at pH 7.5; to obtain pH 2.5, 10 mM KH_2PO_4 was adjusted with HCl.

of strong intermolecular association between PCB5 and PVCL chains. At low-salt conditions, such films are predominantly stabilized by covalent bonds and form high-water-content surface hydrogels.

Figure 5 illustrates that at pH 2.5 PCB5/PVCL-NH_2 hydrogels deswelled in solutions of potassium halide salts. In these conditions PCB5 chains are cationic, and a specific anion binding effect similar to that in Figure 1 is observed. The hydrogel swelling profile in potassium halide solutions was dramatically different at pH 7.5, where hydrogels contained zwitterionic carboxybetaine groups. The observed increased swelling at pH 7.5 reflects typical "antipolyelectrolyte" behavior, or chain expansion in high-salt solutions, which was earlier reported for zwitterionic polymers as indicated by an increase of solution viscosity[52−54] or dynamic light scattering measurements.[55] The chain expansion is caused by disruption of inter- and intramolecular association of positive and negative charges within PCB5 chains[35] and weakening of dipole−dipole interactions between zwitterionic units in solutions with increased salt concentrations. Although this behavior is well-known for polybetaine chains in solutions, here we report that similar salt-induced swelling is observed when polybetaines are included into surface-attached hydrogels. Assuming that all NH_2 groups in PVCL-NH_2 chains are involved in the cross-linking, the (PCB5/PVCL-NH_2)$_5$ surface films are still weakly cross-linked, as they contain ~10 PVCL units between the cross-links, and should allow inclusion of nanometer-size molecules. However, using in situ ATR-FTIR and ellipsometry techniques (data not shown), both in the absence of additional salts and in 0.15 M NaCl solutions at pH 7.4, (PCB5/PVCL-NH_2)$_5$ hydrogels were inert to a variety of negatively charged proteins and biological molecules, such as heparin, IgG, BSA, and DNA. These data are consistent with earlier reported inertness of PBs to biological molecules.[21−23]

In summary, this study describes LbL self-assembly of weak zwitterionic polymers at surfaces driven by hydrogen bonding. The produced films can be erased by lowering ionic strength, switching the counterion type, or increasing the solution pH and are potentially useful for release applications when such films are used as free polyelectrolyte films and separation membranes. The unique feature not previously observed is that counterion type might be used as a trigger to dissolve PCB5/PVCL polymer films. Importantly, hydrogen-bonded films can be chemically cross-linked, producing surface stable films of a new type which showed protein and biomolecule resistant behavior at physiological conditions.

Acknowledgment. This work was supported in part by the National Science Foundation under Award DMR-0513197 and by the American Chemical Society Petroleum Research Fund (Award 41576-AC7). We are grateful to V. Izumrudov for synthesis of PCB5 and many discussions.

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MA071152I