Hydrogen-Bonded Polymer Nanocomposites Containing Discrete Layers of Gold Nanoparticles

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Abstract.

The ability to finely control the spatial location and relative concentration of a nanofiller within a polymer nanocomposite is desirable, especially when faced with fillers that are susceptible to aggregation. In the case of gold nanoparticles (AuNPs), spatial organization is of interest as it offers a means to harness optical and electrical properties, but well-defined placement of AuNPs within a polymer matrix is generally challenging because of phase separation. Here, we demonstrate for the first time the spray-assisted layer-by-layer assembly (LbL) of hydrogen-bonding polymer nanocomposites of poly(ethylene oxide) (PEO) and poly(methacrylic acid) (PMAA) containing discrete regions of AuNPs vertically positioned throughout the film structure. Analysis of the internal structure using cross-sectional transmission electron microscopy (TEM) imaging shows that the AuNP regions are clearly separated by “empty” regions of polymer with no drift or aggregation of the Au NPs during or after fabrication. A UV-Vis spectroscopy study of the stimuli-responsive properties of the spray-assisted LbL nanocomposites shows the release of AuNPs as induced by elevating the environmental pH above the critical pH at which the PEO and PMAA hydrogen bonds are disrupted. We anticipate that this work enables the spatial organization of other nanofillers in stimuli-responsive hydrogen-bonding nanocomposites by spray-assisted LbL assembly.

1. Introduction

Metallic nanoparticles have generated interest because of their unique optical and electrical properties. Specifically, gold nanoparticles (AuNPs) have been utilized for numerous applications including drug-delivery, surface-enhanced Raman spectroscopy, optoelectronics,

biosensing and imaging, and photothermal therapy in cancer treatment.¹⁻⁷ It is often desired to leverage AuNPs by incorporating them into polymer nanocomposites, but aggregation of the nanoparticles is a severe challenge.⁸⁻¹¹ This is often addressed by controlling interactions between the AuNP ligand and the polymer matrix,¹²⁻¹⁵ yet this does not wholly address pre-programmed spatial placement of the NP within the polymer matrix.

Layer-by-layer assembly, a technique by which organic and inorganic multicomponent films are built up on various substrates through complementary interactions of the adsorbing species, offers an opportunity for controlling the spatial placement of nanoparticles vertically within a film with nanometer-scale precision.¹⁶⁻¹⁸ Complementary interactions between species can be electrostatic (alternating between positively and negatively charged species) or secondary forces like Van der Waals and hydrogen bonding. The species used to build up the film do not have to be explicitly polymers and can include biological molecules and inorganic nanoscale fillers for a range of applications.^{1,3,19-25} For example, AuNPs have been assembled into LbL nanocomposites films to demonstrate tunable conductivity,^{26,27} sensing platforms,²⁸ drug delivery,^{2,22,24} and optoelectronics.²⁹ The vast majority of these studies have dealt with the LbL assembly of AuNPs by electrostatic interactions and by a form of LbL assembly called “dip-assisted” LbL assembly, which relies on total immersion of the substrate.^{20,23,30-32}

Fewer studies have been dealt with hydrogen-bonded LbL systems with AuNPs, which are interesting alternatives to their electrostatic counterparts. The pH sensitivity of hydrogen-bonded multilayer systems renders them stimuli-responsive with tunable, switchable properties for potential use in biomedical applications such as drug delivery systems.^{22,24,25,33} The ability to add non-charged molecules or particles to the hydrogen-bonded composite structure is also of interest for sensor-type or delivery-type systems that utilize changes in environmental conditions to

release pay-loads through controlled dissolution of the LbL structure.²² In one example, the Tsukruk group reported the reversible controlled distribution of gold nanorods (AuNRs) within hydrogen-bonded poly(*N*-vinylpyrrolidone)(PVPON)/PMAA multilayer films by changing the pH of the environment, which altered the optical properties of the films.³⁴

AuNP-containing multilayer films have been prepared by dip-assisted and spin-assisted LbL assembly methods.^{26,28,34} Dip-assisted LbL assembly is based on the immersion of the substrate, whereas spin-assisted LbL assembly is based on spin-coating. Both dip-assisted and spin-assisted LbL assembly approaches have drawbacks. Dip-assisted LbL assembly requires relatively long adsorption times, on the order of 1-15 minutes per layer. On the other hand, spin-assisted LbL assembly is much more rapid, on the order of seconds per layer; however, this approach is limited to flat substrates. The spray-assisted LbL technique is perhaps more suitable, where adsorbing species are alternately sprayed onto a substrate. This requires only a few seconds of exposure and is applicable to a wide range of substrate materials with varying geometries.^{27,35-38} Negatively charged AuNPs have been assembled with a polycation by spray-assisted LbL assembly successfully using electrostatic interactions.³⁹ However, AuNP-containing hydrogen-bonded multilayer films made by spray-assisted LbL assembly have not yet been reported in the literature.

Here, we report hydrogen-bonded multilayers made by dip-assisted and spray-assisted LbL assembly in which AuNP-rich layers are placed at predetermined depths within the film. PEO/PMAA was selected as the model hydrogen-bonded system,^{19,22,35,40,41} for which PEO acts as the hydrogen bond acceptor and PMAA acts as the hydrogen bond donor. This system is pH-responsive, having a reported critical pH of 4.5 above which hydrogen bonds become disrupted.⁴⁰ AuNPs were selected as the nanoscale filler because of their optoelectronic

properties, which allow us to track aggregation and LbL dis-assembly. The layered nanocomposite structure was verified using TEM, and the distance between the AuNP-rich layers was varied by altering the number of PEO/PMAA layer pairs between AuNP exposures. UV-Vis spectroscopy was used to determine AuNP aggregation (or lack thereof) as well as the time-dependent release of AuNPs from a film exposed to an environmental pH above the critical pH. We hypothesized that incorporating AuNPs into pH-sensitive systems like PEO/PMAA at predetermined depths separated by predictable, variable distances should result in tailorable delivery timescales. This investigation lends a fundamental understanding of the resulting structure of LbL nanocomposites upon addition of AuNPs to PEO/PMAA LbL films using dip- and spray-assisted LbL assembly.

2. Experimental

2.1. Materials

Poly(ethylene oxide) (PEO) ($M_w = 4,000,000$ g/mol and $M_w = 100,000$ g/mol, Sigma Aldrich) and poly(methacrylic acid) (PMAA) ($M_w = 100,000$ g/mol, Polysciences) were used as received. Chlorauric acid (99.999% trace metal basis, Sigma Aldrich) and trisodium citrate dihydrate (Sigma Aldrich) were used to prepare the AuNP colloidal suspension. Linear poly(ethyleneimine) (LPEI) ($M_w = 25,000$ g/mol, Polysciences) and poly(acrylic acid) (PAA) ($M_w = 50,000$, Polysciences) were used as received. For the LbL assembly processes, the PEO and PMAA solutions were prepared with a concentration of 20 mM in Milli-Q water based on repeat unit, and the pH was adjusted to 4 using HCl and NaOH stock solutions. Indium tin oxide (ITO) coated glass slides (Delta Industries) were used as substrates for the LbL assembly

procedures. Poly(glycidyl methacrylate) (PGMA, $M_w = 25,000$ g/mol, Polysciences) was used as-purchased for surface treatment of the substrates.

2.2. Preparation of AuNP colloidal suspension

The AuNP suspension was prepared using the traditional Turkevich Method.^{27,42} In short, chlorauric acid (1 mM in water) was brought to a boil using a hot plate. A 1 wt% solution of trisodium citrate dihydrate in water was added to the boiling chlorauric acid solution and stirred vigorously for 15 min then removed and allowed to cool to ambient temperature. The solution turned from yellow to clear to black to a deep red color, indicating the completion of the AuNP formation process. The AuNP suspension was kept refrigerated away from light until use to prevent aggregation. The resulting AuNPs were examined using transmission electron microscopy (TEM), and the average diameter was determined to be roughly 15-20 nm.

2.3. Substrate preparation for LbL film fabrication

ITO-coated glass substrates were cleaned *via* sonication in 0.5 M NaOH for 10 min followed by rinsing with Milli-Q water and blow-drying with nitrogen gas. The substrates were then immersed in a water/NH₄OH/H₂O₂ (5:1:1 by volume) solution at 75°C for 20 min followed by rinsing three times and sonication in Milli-Q water for 5 min three times. After the rinsing step, the substrates were blow-dried and treated with O₂ plasma for 10 min under vacuum. Next, the substrates were immersed in 0.1% PGMA in methyl ethyl ketone (MEK) solution for 3 min followed by annealing in a convection oven at 110°C for 30 min.³⁵ After annealing, the substrates were rinsed with MEK 3 times at room temperature. Finally, the substrates were immersed in a PMAA (20 mM in water) solution at room temperature for 20 min followed by annealing at 110°C for 30 min and a final rinsing step with Milli-Q water.

2.4. Substrate preparation for pH-modulated release study

ITO-coated glass substrates were exposed to O₂ plasma and PGMA surface treatments as previously described. Then, 4 layer pairs of LPEI and PAA were deposited as anchor layers to initiate film growth. The LPEI/PAA initial layers were used to initiate film growth because a substantially thick initial layer of PEO/PMAA was not necessary as it would not influence the absorbance spectrum of the films during dissolution and release of the AuNPs.

2.5. Substrate preparation for cross-sectional TEM analysis

For TEM analysis Dupont Tiejin ST505 7-mil PET was used as the substrate material. PET was cleaned by rinsing with methanol and DI water (4x). They were then exposed to O₂-plasma for 10 min prior to LbL assembly, and PGMA treatment was not conducted.

2.6. Dip-assisted layer-by-layer assembly of hydrogen-bonded PEO/PMAA films and PEO/PMAA nanocomposite films containing AuNPs

PEO/PMAA films were fabricated using a programmable slide-stainer (HMS Series, Carl Zeiss, Inc.). Treated ITO-coated glass substrates were immersed in the PEO solution (20 mM in Milli-Q water by repeat unit) for 15 min followed by 3 consecutive rinsing steps in deionized water baths for 2, 1, and 1 min. The substrates were then immersed in the PMAA solution (20 mM in Milli-Q water by repeat unit), for 15 min followed by the same rinsing pattern. The pH was adjusted to 4 for all solutions. This cycle constitutes a layer pair, and was repeated n times. Hence, LbL films of PEO/PMAA are denoted (PEO/PMAA) _{n} where n is the number of layer pair cycles.

AuNP-containing nanocomposite films were prepared using similar methods as described for (PEO/PMAA)_n films with some modification to the procedure. Periodically, PEO/AuNP layers were deposited by hand instead of PEO/PMAA layers. To do this, the films were immersed in PEO solution for 15 min followed by rinsing in deionized water for 2, 1, and 1 min. The films were subsequently immersed in AuNP suspension for 14 min followed by the same rinsing steps (following prior literature²⁷). The pH was adjusted to 4 for all solutions.

2.7. *Spray-assisted layer-by-layer assembly*

As in the dip-assisted LbL procedure just described, identical solutions of the same concentration (20 mM) as well as pH (4) were used for spray-assisted LbL assembly of the (PEO/PMAA)_n films. Films prepared on ITO-coated glass and Dupont Tiejin ST505 7-mil PET (for TEM imaging) were cleaned and exposed to O₂-plasma as mentioned earlier. The PET substrates were not subjected to PGMA surface treatment because the required annealing temperature was too high. The films were assembled using an automated spraying system (Svaya Nanotechnologies). All substrates used for spray-assisted LbL assembly were placed roughly 20 cm away from the spray nozzles, and the solutions were sprayed at a pressure of about 40 psi. First, the PEO solution was sprayed on the substrate for 10 s followed by 10 s of draining and 10 s of rinsing with Milli-Q water with a 10 s pause to allow excess solution to drain between layer pair cycles. The PMAA solution was then sprayed following the same pattern. This deposition cycle was repeated until the desired number of layer pairs (*n*) were deposited on the surface.

Nanocomposite films containing AuNPs were prepared in a similar fashion to the AuNP-free films. The assembly pH was adjusted to 4 for all solutions as well as the AuNP suspension. Initial layers of PEO/PMAA (typically around 50 layer pairs) were deposited as base layers in order to ensure substantial growth behavior of the film. We found that films produced using

fewer initial PEO/PMAA layer pairs were much thinner than those produced with a more substantial base layer of PEO/PMAA. Once the base layers were complete, alternating layers of PEO solution and AuNP suspension were deposited using the same timing parameters as mentioned in the previous section until the desired number of PEO/AuNP layer pairs (m) was achieved. Then, a predetermined number of PEO/PMAA layer pairs was deposited in order to separate the previous (PEO/AuNP) $_m$ region from the next one. This spraying sequence was repeated until the desired amount of (PEO/AuNP) $_m$ regions was achieved. The film was then capped with a final layer of (PEO/PMAA) $_s$, where s varied from 10-20 layer pairs.

For the pH-responsive study, nanocomposite films were prepared *via* spray-assisted LbL assembly. In order to facilitate initial growth of the film, anchor layers of (LPEI/PAA) $_4$ were deposited onto the substrate. The (LPEI/PAA) $_4$ anchor layers were used in place of (PEO/PMAA) $_{50}$ for these films in order to expedite film fabrication time while still achieving a good foundational layer for quality film growth. All layer pairs were sprayed using the same standard parameters listed previously. Subsequently, (PEO/AuNP) $_m$ regions, where m denotes the number of layer pairs, and separator (PEO/PMAA) $_n$ regions, where n was either 2 or 12 layer pairs, were deposited to build up the nanocomposite containing discrete regions of AuNPs. Once the desired film composition was achieved, the films were capped with (PEO/PMAA) $_{10}$.

2.8. TEM analysis

Cross-sectional TEM imaging was done using a TECNAI F20 Super-Twin transmission electron microscope fitted with a Schottky field emission gun, a $2k \times 2k$ Gatan CCD camera. Distances between AuNP-rich regions of the composite films as well as AuNP sizes were calculated using Digital MicrographTM software. Dimensions of the films were calculated using

scale bars integrated into each image. The nanocomposite LbL films for TEM analysis were prepared in a similar manner to those built upon ITO-coated glass substrates, but Dupont Tiejin ST505 7-mil PET was used as the substrate material instead. The TEM substrates were cleaned by rinsing with methanol and DI water (4x). They were then exposed to O₂-plasma for 10 min prior to LbL assembly. Samples prepared on PET substrates were cut down to 1 mm × 1.5 cm strips and embedded in epoxy using a silicon mold. They were then placed in a vacuum chamber until air pockets were sufficiently removed from the epoxy. The epoxy-embedded substrates were then held at 40°C overnight. Finally, the samples were trimmed to the appropriate size *via* microtoming with care not to damage the LbL film for imaging.

2.9. UV-Vis spectroscopy

UV-Vis spectroscopy was conducted using a Shimadzu SolidSpec-3700 UV-Vis-NIR spectrophotometer. For the AuNP-release study, dry AuNP-containing nanocomposite films on ITO-glass substrates were placed immersed in water and the pH was adjusted in order to induce disruption of the hydrogen bonds. Aliquots of the solution were taken and the absorbance was measured at selected times throughout the experiment in order to monitor the breakdown of the films. After each measurement, the aliquot sample was returned to the film environment to avoid concentration changes that would accompany change in volume. The final pH generally decreased, but remained well above pH 4.5.

2.10. Profilometry

Thickness was measured using profilometry (Alphastep D-100, KLA-Tencor). Measurements were taken in at least three different locations across each sample to obtain an average thickness.

3. Results and Discussion

3.1. Cross-sectional TEM analysis

Figure 1. (a) Chemical structures of polymers used in the hydrogen-bonded system and illustration of the structure of the citrate-stabilized AuNPs used as nanoscale filler (not drawn to scale) and (b) an illustration of the cross section of an AuNP-containing LbL composite film.

Figure 1a shows an illustration of the citrate-stabilized AuNP. For simplicity, only the citrate ligands are shown on the AuNP surface, although it has been shown that some additional oxidation products can form on the surface during the synthetic process.^{43,44} In our study, we assume that any effects these impurities may cause are minimal and have no impact on the results of the study. Figure 1b show an LbL assembled nanocomposite films containing discrete regions of AuNPs throughout the film structure. We were able to achieve this type of film architecture using spray-assisted and dip-assisted LbL assembly. The structure of the

$(\text{PEO/PMAA})_n/[(\text{PEO/AuNP})_m/(\text{PEO/PMAA})_p]_x/(\text{PEO/PMAA})_s$ nanocomposites film- where n , m , p , x , and s represent the number of layer pairs making up that region of the film - consisted of base layers to facilitate growth, intermediate layers where there were discrete regions of AuNPs, and cap layers to facilitate TEM imaging and cargo-release studies.

Figure 2 (a-b) shows cross-sectional TEM images of dip-assisted nanocomposite films prepared at pH 4 with and without $(\text{PEO/AuNP})_m$ layers for comparison. The pH was adjusted to 4 in order to keep the AuNPs from flocculating and also to stay below the pH_{crit} of the $(\text{PEO/PMAA})_n$ system. In this manner, the entire assembly process proceeded by hydrogen bonding interactions. The regions containing the AuNPs consisted of $(\text{PEO/AuNPs})_1$ separated by $(\text{PEO/PMAA})_2$. It can be seen in the image that the early stages of the film growth show a higher concentration of AuNPs closer to the substrate, with a clear decrease in relative concentration as the number of PEO/AuNPs deposition cycles increased. This was attributed to aggregation of the AuNPs within the reservoir bath used for the dip-assisted LbL process. The aggregation was likely due to loosely bound polymer chains detaching from the substrate into the AuNP suspension during the deposition step (i.e. cross-contamination). Visual verification of the aggregation could be seen in the color change of the AuNP suspension from deep red to a nearly black color throughout the course of the experiment. Cloudy flocculation of the AuNPs could also be seen in the solution after only a few deposition cycles.

Figure 2. TEM images of (a) dip-assisted LbL PEO/PMAA films containing no AuNPs, (b) dip-assisted LbL nanocomposite containing several $(\text{PEO}/\text{AuNPs})_1$ regions separated by $(\text{PEO}/\text{PMAA})_2$, (c) spray-assisted LbL PEO/PMAA nanocomposite consisting of several $(\text{PEO}/\text{AuNPs})_3$ regions separated by $(\text{PEO}/\text{PMAA})_{12}$, (d) high-magnification image showing relative sizes AuNPs within the film prepared by spray-assisted LbL assembly. The labels in panels b-c correspond to average distance between Au-rich regions.

In order to eliminate the issue of AuNP aggregation during film buildup, the spray-assisted LbL technique was employed. Using this technique, the possibility of aggregation was minimized because the substrates and polymer chains loosely bound to the film surface no longer directly contacted the AuNP suspension. Figure 2 (c) shows a cross-sectional TEM image of one of the nanocomposite films. AuNP-rich regions of the film consisting of $(\text{PEO}/\text{AuNPs})_3$ were

separated by (PEO/PMAA)₁₂. These TEM images show the successful inclusion of distinct, discrete regions of AuNPs separated by “empty” AuNP-free regions. Based on these images, the observed individual layer pair thickness for the PEO/PMAA regions of the spray-assisted LbL film was significantly thinner than that of the dip-assisted regions in which the (PEO/AuNP)₁ region was separated by (PEO/PMAA)₂. Specifically, the separation between Au-rich regions was 400 nm for (PEO/PMMA)₂ by dipping and around 300 nm for (PEO/PMAA)₁₂ by spray. This was expected, as previous work demonstrates that spray-assisted LbL generally results in thinner layers.³⁵ Spray-assisted LbL also affords significantly faster layer-pair deposition times. For films of identical composition, the time scale for sample production can drop from several days to a few hours or minutes, depending on the desired number of layer pairs being deposited. Further, we calculated that spray-assisted LbL assembly consumed much less polyelectrolyte solution as compared to dip-assisted LbL assembly (4 mL vs 10 ml polyelectrolyte solution/layer pair, respectively).

To examine the effect of the number of PEO/PMAA separation layers (denoted as p in Figure 1) in spray-assisted LbL films, two films were prepared at $p = 2$ and 12 layer pairs. As p increases, the distance between AuNP-rich regions should increase. All other layer pair numbers were fixed as: (PEO/PMAA)₁₅/[(PEO/AuNPs)₃/(PEO/PMAA) _{p}]₅₀/(PEO/PMAA)₁₀. For comparison, an AuNP-free LbL film composition of (PEO/PMAA)₅₂₀ was prepared, which would be analogous to a case where $p = 12$ layer pairs. The films produced with (PEO/PMAA)₂ separation layers were significantly thinner than those with separation layers of (PEO/PMAA)₁₂ (530 nm vs 4.3 μ m, respectively). The thickness of the AuNP-free film was 4.9 μ m, slightly higher than the $p = 12$ AuNP-containing case. These results indicate indirectly that the separation distance can be tuned by adjusting the number of PEO/PMAA layer pairs p between AuNP-rich

regions. This demonstrates that a high degree of control over the placement, spacing, and relative concentration per AuNP-rich region can be achieved through changing the number of PEO/PMAA layer pairs separating the AuNP-rich regions or by adjusting the number of PEO/AuNP layer pairs in each of those regions, respectively.

3.2. Optical microscopy

The surface morphology of the PEO/PMAA nanocomposites containing AuNPs was examined using optical microscopy (see Figure 3). The films and nanocomposites produced by spray-assisted LbL assembly had smooth surfaces with few defects, similar to previous reports for spray-assisted LbL films consisting of PEO/PMAA.³⁵ Previous reports on the surface morphology of dip-assisted LbL films consisting of PEO/PMAA attribute roughness on the surface to the hydrophobic interactions between the α -methyl groups on the PMAA backbone.⁴⁵ Similar surface roughness behavior was observed by Seo et al. where LbL assemblies of hydrophobically modified PEO and poly(acrylic acid) (PAA) showed large surface grains resulting from PEO micelle formation.⁴⁶ In contrast, the smoother morphological nature of spray-assisted LbL films observed here could be due to insufficient time for chain penetration and rearrangement during assembly. Additionally, the shorter contact time afforded by spray-assisted LbL assembly combined with the possibility of chain spreading during the spray process could be responsible for the smooth surface morphology of these films.⁴⁷ Figure 3 (a) and (b) show some visible features on the surface while (c) shows none. These features are likely caused by phase-separated regions of PEO and PMAA. The lack of these features on the surface of the LbL nanocomposites with $p = 2$ separation layers could be indicative of the effect of the AuNPs on the interactions between PEO and PMAA. It should also be noted that the LbL nanocomposites with $p = 2$ separation layers were significantly thinner (less than one micron) than the NP-free

and the nanocomposites with $p = 12$ separation layers. These thin nanocomposites consist of significantly less polymer while the number of AuNP layers deposited for both $p = 12$ and $p = 2$ nanocomposites were kept the same.

Figure 3. Optical microscopy images showing the surface morphology of (a) PEO/PMAA AuNP-free (b) PEO/PMAA nanocomposite with $p = 12$ and (c) PEO/PMAA nanocomposite with

$p = 2$ LbL films produced by spray-assisted LbL assembly. Images taken at 20x magnification using reflected light. All samples have capping layers of (PEO/PMAA)₁₀.

3.3. UV-Vis spectroscopy

Spray-assisted PEO/PMAA LbL films with and without AuNPs were analyzed using UV-Vis spectroscopy. Figure 4 shows UV-Vis spectra for films of separation layers $p = 2$ and 12 and of (PEO/AuNPs)₃. These films were designed such that the number of AuNP layers are equal for both $p = 2$ and 12. Notably, the peak absorbance values were similar for both $p = 2$ and 12, suggesting that these two samples have a similar number of AuNPs. Further, the peak positions are similar at 530 nm, indicating that the AuNPs have similar environments. No significant red-shift in the spectrum was observed, as compared to an AuNP suspension, implying that there was no major aggregation of AuNPs within the film matrix during or after the film deposition process. However, the slight increase in absorption in the 600-900 nm range of the spectra for the nanocomposite films compared to the pure AuNP solution is likely the result of some lateral aggregation within the nanocomposite.⁴⁸ This suggests that some small amount aggregation may occur locally within the segregated regions of the film containing AuNPs, causing the slight absorption increase at the higher wavelengths.⁴⁹ No significant aggregation of the nanofiller at the surface or within the film implies that spray-assisted LbL provides good control over the dispersion of the AuNPs across the surface of the film during deposition.

Figure 4. (a) UV-Vis spectra showing the absorbance of LbL films fabricated by spray-assisted LbL assembly with and without AuNPs and the as-prepared AuNP suspension. The film compositions presented are (PEO/PMAA)₁₅/[(PEO/AuNPs)₃/(PEO/PMAA)_{*p*}]₅₀/(PEO/PMAA)₁₀ where *p* was 2 (red) or 12 (blue) and (PEO/PMAA)₅₂₀. (b-c) Digital images of a PEO/PMAA film containing discrete regions of AuNPs and pure (PEO/PMAA)₅₂₀ film containing no AuNPs, respectively.

3.4. pH-responsive gold release study

The pH-responsive behavior of the nanocomposite films was next explored. This was motivated by the reported pH-responsive properties of the hydrogen-bonding PEO/PMAA system (without AuNPs).^{40,41} Previously, the pH-driven deconstruction of PEO/PMAA films was demonstrated by the release of a small molecule dye (Rhodamine 6G) embedded in the film upon exposure to an environmental pH above the pH_{crit} of the polymer film (4.5). At a slightly elevated pH of 5.5, hydrogen bonds were disrupted because of the increase in the ionization of carboxylic acid pendant groups along the PMAA chain. This disruption was followed by electrostatic repulsion among the (COO⁻) groups and a change in the physical conformation of the polymer from a more coiled structure to a more extended structure. The breaking of the

hydrogen bonds coupled with the change in chain conformation resulted in the dissolution of the film, and release of the dye contained therein.⁵⁰ Here, we demonstrate a pH-responsive, time-dependent release of AuNPs from PEO/PMAA nanocomposites containing AuNPs fabricated using spray-assisted LbL assembly.

Nanocomposite films were prepared *via* spray-assisted LbL assembly with the composition (LPEI/PAA)₄/[(PEO/AuNP)₃/(PEO/PMAA)₁₂]₅₀/(PEO/PMAA)₁₀. Dried samples were immersed in 10 mL of water at pH 10 and pH 11 - well above the reported pH_{crit} of the hydrogen bonded system⁵⁰ - and stirred vigorously using a small stir bar. As described earlier, the elevated pH induced the breakdown of the LbL nanocomposite structure through disruption of the hydrogen bonds. This breakdown resulted in the release of the AuNPs into the local environment. The release of AuNPs was monitored by retrieving aliquots from the solution and collecting UV-Vis spectra over the course of several hours (see Figure 5).

Figure 5. (a-b) UV-Vis data collected from a payload release experiment on AuNP-containing nanocomposite films exposed to pH 10 and 11 solutions, respectively, and (c) normalized absorbance with respect to time showing the progression of release of the AuNP payload at both pH levels at a wavelength of 525 nm. (d) An illustration of LbL dis-assembly and release of AuNPs. All films were made by spray-assisted LbL assembly.

As expected, the release of the AuNPs took place faster at pH 11 than at pH 10 because of the greater driving force for film deconstruction. In both cases, the nanocomposites completely dissociated into solution, releasing their entire payload. This is denoted by a steady increase in the peak at 525 nm, attributed to the AuNPs being released into the surrounding solution. Although the data above demonstrates successful erasability of the nanocomposite film and time-dependent delivery of the AuNP payload, the time scale of the film dissolution was generally longer (on the scale of hours) than expected. It has been shown by the Sukhishvili group that hydrogen bonded films released a dye payload rapidly (on the scale of seconds) upon increasing the pH above the pH_{crit} of the system, but those results were for the release of small molecules, not large nanoparticles, as in our case.⁵⁰ Many payload delivery systems reported in the literature deal specifically with single component hydrogels or cross-linked networks that respond to environmental stimuli and rely on swelling and deswelling and diffusion of the payload from the matrix for delivery.⁵¹⁻⁵⁴ The differences in the time scale of the release are most likely connected to the large size and mass of the AuNP as compared to that of a small molecule.

We hypothesize that the brief study here shows release of the AuNPs by simple diffusion from the surface of the film as the dissolution takes place. Since no crosslinking was induced by post processing methods, it is likely the film dissolution of the PEO/PMAA layers occurred rapidly as expected. However, the limiting factor involved in the release of the AuNPs could possibly be

attributed to favorable interactions (i.e. hydrogen bonding and Van der Waals forces) between the PEO and citrate ligands on the AuNP surfaces. The relatively high surface area of the 15-20 nm diameter particles provides good contact between the AuNPs and the surrounding polymer chains, which could lead to slower diffusion from the surface despite the higher pH of the environment causing disruption of the hydrogen bonds. This may also to possible ligand exchange and chelation with ionized PMAA groups that may also hinder the release. One might have expected a burst-type release from the film, had there been steady surface erosion, but this was not observed. Perhaps this might be attained by increasing the p , the number of layer pairs separating the AuNP-rich regions. In general, the AuNP release from PEO/PMAA LbL assemblies demonstrates the possibility of delivering or controlling payload release using a pH trigger.

4. Conclusion

A highly efficient and well-organized method of polymer nanocomposite fabrication was utilized to produce hydrogen-bonded layer-by-layer films containing discrete regions of gold nanoparticles. The hydrogen-bonding polymers used in this study were utilized as a vehicle for time-dependent release of the gold nanoparticles when exposed to pH conditions sufficient to disrupt the secondary bonding. These results verified our hypotheses that spray-assisted LbL assembly could be leveraged to controllably place AuNPs through the thickness of the thin film and that disruption of the hydrogen-bonding interactions could lead to the programmed disassembly of the film and AuNP release.

The high degree of control over the internal structure and placement of the gold nanoparticles provided by the spray-assisted layer-by-layer assembly method offers a vast improvement over alternative nanocomposite fabrication methods such as drop-casting, spin-casting, and dip-

coating in terms of fabrication time, uniformity of filler distribution, and minimization of aggregation.^{36,38,39,55} These methods often require additional, more extensive surface treatment of the nanoparticles such as ligand exchange or polymer grafting to prevent aggregation within the cast nanocomposite films.⁵⁶⁻⁵⁸ The scalability of the spray-assisted LbL technique combined with the extensive control over the internal structure and properties of resulting films and nanocomposites is a valuable feature that has the potential to produce novel materials for specialized applications that could otherwise not be achieved.⁵⁹ Polymer nanocomposites formed using spray-assisted layer-by-layer assembly could potentially be used as drug delivery systems or incorporated into responsive membranes or materials where external stimuli would activate the delivery process.^{21,32,60} To this end, with the approach shown here, the release properties of the films can be altered easily through changing the number of layer pairs separating the nanoparticle regions or by modulating the environmental pH conditions. Our future vision includes translating this approach to other colloidal nanoparticles and polyelectrolyte systems to demonstrate that this approach may be broadly suitable for many other colloidal materials and polymer systems beyond just AuNPs. This would, in turn, show precise vertical placement of a broad range of nanomaterials within a well-defined film for numerous applications.^{22,26,35,61}

Author Information

Author Contributions

The manuscript was written through contributions of all authors. All authors have given approval to the final version of the manuscript.

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