

Article

SERS and Chiral Properties of Cinnamic Acid Derivative Langmuir-Blodgett Films Complexed with Dyes

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Abstract

Chiral molecules are crucial in the field of optical devices, molecular recognition, and other novel functional materials due to their unique spatially asymmetric configuration and optical activity. In this study, a chiral molecule, Cholest-3-yl (E)-3-(4-carbamoylphenyl)acrylate (CCA), was combined with dyes containing large conjugated structures, tetramethylporphyrin tetrasulfonic acid (TPPS), and Nickel(II) phthalocyanine-tetrasulfonic acid tetrasodium salt (TsNiPc), and composite LB films of CCA/TPPS and CCA/TsNiPc were successfully prepared by using Langmuir-Blodgett (LB) technology. The circular dichroism (CD) test proved that the CCA/TPPS composite film had a strong CD signal at 300–400 nm, and the composite film showed chirality. This significant optical activity provides a new idea and option for the application of LB films in chiral sensors. In the Surface Enhanced Raman Spectroscopy (SERS) test, the CCA/TPPS composite film was sensitive to signal sensing, in which the enhancement factor $EF = 2.28 \times 10^5$, indicating that a large number of effective signal response regions were formed on the surface of the film, and the relative standard deviation (RSD) = 12.08%, which demonstrated that the film had excellent uniformity and reproducibility. The high sensitivity and low signal fluctuation make the CCA/TPPS composite LB film a promising SERS substrate material.

Keywords: chiral; Langmuir-Blodgett; SERS; sensor



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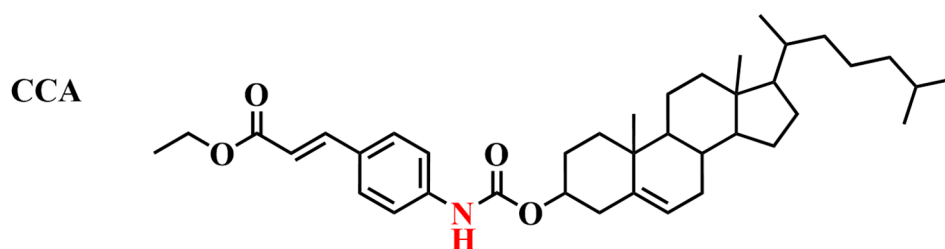
1. Introduction

Chirality is a common phenomenon in organic compounds, extensively utilized in optics, chemical synthesis, and other areas due to its structural features of left-right spin asymmetry [1–4]. As a notable aromatic organic acid, cinnamic acid has attracted significant interest from scientists recently because of its excellent photoresponsivity, high biocompatibility, and eco-friendliness. The benzene ring in cinnamic acid can be replaced with other groups, such as hydroxyl and methoxy, to produce a variety of derivatives with broad applications across multiple industries. The outstanding biocompatibility and strong antioxidant properties of phenylacetic acid have generated widespread interest, leading to the use of cinnamic acid derivatives in various medicinal fields, including anticancer, antioxidant, and antibacterial therapies [5–12]. Additionally, cinnamic acid groups can form nano-enzymatic materials for biosensing or antimicrobial applications by acting as ligands with transition metals [13,14]. The application of click chemistry in functional materials

has generated cinnamic acid derivatives with enhanced optical properties. The double bond in acrylic acid can dimerize under UV light to form a cyclobutane structure, which is useful in adhesives, smart coatings, light-curing hydrogels, and photoresponsive materials [15–22]. Takada studied the photodeformation behavior of the synthesized cinnamic acid-based polymer under UV light and observed that the PdHCA surface exhibits photo contraction, whereas the P3HCA surface shows a unique photo expansion behavior [22,23]. The excellent biomaterial properties and photoresponsive properties of the cinnamic acid group have attracted the attention of researchers, yet there are few applications for trace substance detection and chiral sensors.

Self-assembled thin film technology is a thin film preparation technique that spontaneously creates ordered structures through intermolecular interactions, which has given rise to important advancements in material design and performance optimization. Asymmetric self-assembled monolayers (SAMs) are a surface modification technique that depends on the spontaneous adsorption of molecules on the substrate surface to form a monolayer. The precise interfacial regulation ability and wide material compatibility make them useful in the energy, electronics, and biosensing industries [24–27]. However, molecular accumulation causes SAMs to be prone to uneven film formation, which impacts device performance. Langmuir-Blodgett (LB) technology is an advanced method for preparing monomolecular films that allows the transfer of layers formed by self-assembly at the gas-liquid interface onto solid substrates under specific conditions, while maintaining interfacial order. Because the LB technique uses the Langmuir tank to precisely control the transfer rate and surface pressure, the resulting films are more uniform. In LB technology, vertical lift and horizontal transfer methods are commonly used to move monolayers from the gas-liquid interface to a solid base. The vertical lifting method involves immersing the solid substrate vertically into the subphase solution containing spreading molecules. Once a stable monomolecular layer forms at the interface, it is transferred to the substrate by lifting or lowering the base at a uniform speed in a controlled manner. The horizontal transfer method involves bringing the solid substrate into contact with the molecule layer at the interface, enabling direct transfer of the layer to the substrate surface through intermolecular forces. Nanomaterials' high specific surface area and superior optical and chemical characteristics give them distinct benefits in LB technology [28–31]. For example, Almeida [28] has made a layered non-metallic g-C₃N₄ LB film that performs excellently in chemical conversion, photovoltaics, and photocatalysis. However, the organic materials, particularly those that combine with large π -conjugated structures forming LB films, have not been deeply investigated. In light of this, the goal of this work is to create composite LB films, which combine organic molecules and massive π -conjugated systems. The Cholest-3-yl (E)-3-(4-carbamoylphenyl)acrylate (CCA) molecule (Scheme 1) is a chiral-containing cinnamic acid derivative. Tetraphenylporphyrin sulfonate (TPPS) is a porphyrin derivative with a highly conjugated π -electron system. Its unique anionic nature allows it to bind easily to cations, making it useful for detecting heavy metals in environmental or biological samples. Nickel(II) phthalocyanine-tetrasulfonic acid tetrasodium salt (TsNiPc) is extensively used in sensing technology and environmental treatment because of its tunable electronic properties. Surface-enhanced Raman spectroscopy (SERS) is commonly employed for trace marker detection in biochemistry and environmental monitoring due to its ultra-high sensitivity [32–37]. Molecular assembly at the nanometer level can significantly increase contact areas and enhance reaction sensitivity. Therefore, LB films that combine CCA molecules with TPPS or TsNiPc have the potential to serve as highly sensitive SERS substrates with excellent signal reproducibility. Chiral materials exhibit significant effects in physical, chemical, and optical properties because of the absence of mirror symmetry and are widely used in fields such as optoelectronic materials and information encryption [38–42]. The LB

technique can manipulate the interfacial aggregation of chiral molecules, thereby regulating their chiral optical properties. Studying the chiral characteristics of CCA molecules within composite LB films can provide new insights for developing novel chiral materials.



Scheme 1. Schematic molecular structures of CCA.

2. Experimental Section

2.1. Materials

CCA molecules were synthesized according to related reports [23]. Trichloromethane (TCM), Hydrochloric acid (HCl), Rhodamine 6G (R6G), TPPS, and TsNiPc reagents were purchased from Aladdin Reagent & Chemical Company, Shanghai, China. Ultrapure water was prepared from UPY-II-103.

2.2. Preparation of Films

CCA/TPPS and CCA/TsNiPc composite LB films were prepared at room temperature using a KSV-NIMA Langmuir film analyzer. To ensure a clean experimental environment, the LB tank was cleaned with alcohol and ultrapure water, which is water with a resistivity of $18.2 \text{ M}\Omega\cdot\text{cm}$ (25°C) or more, before the experiment. The pH of the aqueous solution was adjusted to 3–4 with concentrated hydrochloric acid. Then, appropriate amounts of TPPS and TsNiPc powders were weighed and dissolved in the aqueous solution to prepare a subphase solution with a concentration of $1 \times 10^{-4} \text{ mol/L}$. Moreover, the CCA molecules were immersed in the TCM solution with a concentration of 0.1 mg/mL . During the experiments, 250 mL of the subphase was slowly poured into the LB tank at room temperature and pressure. Then, $100 \mu\text{L}$ of CCA solution was slowly and evenly dripped onto the surface of the subphase using a microsyringe. After 20 min, the CCA molecules were fully bound to the subphase. The sliding barriers on both sides of the LB tank were gradually compressed on the surface of the subphase at a speed of 8 mm/min . The surface pressure of the subphase was measured in real time by a tensiometer attached to the Wilhelmy plate, from which a π -A curve was obtained. Once the desired pressure (20 mN/m) was reached, the monolayer or multilayer LB films were transferred to a solid substrate—such as a carbon support film, mica sheet, quartz sheet, or glass sheet—using either vertical lifting or horizontal transfer methods for further morphology characterization and property studies. During this process, 200 mL of the subphase was slowly poured into the LB tank at room temperature and pressure.

3. Characterization

To obtain morphology data on the composite film, the monolayer was transferred onto a carbon-supported surface utilizing the horizontal transfer method for transmission electron microscopy (HT 7700, Tokyo, Japan) testing. The vertical lift-off method was used to transfer the monolayer film onto a stripped mica sheet. Atomic force microscopy (AFM) was used to test the composite film using a Nanoscope Multimode 8 scanning probe (MA, USA). The horizontal transfer method was used to transfer 60 layers of films onto quartz sheets, and tests were conducted using the UV-vis (UV-2550, Shimadzu, Japan), contact

angle (SDC-350, Dongguan, China), and circular dichroism (J-810, Tokyo, Japan) techniques. The multilayer film was moved to a glass sheet using the horizontal transfer method, and before the test, the sample was sliced into 5×5 mm pieces for SERS (London, UK) testing.

4. Results and Discussion

The surface pressure-area isotherm (π -A) reflects phase transitions, molecule arrangements, and film stability by tracking surface pressure (π) as a function of molecule area (A) in monolayers. The increase of surface pressure is directly related to the enhancement of intermolecular forces and the improvement of the degree of ordering of molecular arrangement. The increase of intermolecular force and the improvement of molecular ordering degree led to the decrease of the average area occupied by molecules and the increase of surface pressure of the composite film. The pure TPPS and pure TsNiPc solutions, CCA molecules' π -A isothermal curves on ultrapure water, TPPS, and TsNiPc subphases are displayed in Figure 1. When the molecular area approaches $1.12 \text{ nm}^2/\text{molecule}$ in the pure TPPS solution, the surface pressure of the composite film begins to increase gradually. It then increases as the single-molecule film is compressed further, eventually reaching a collapse pressure of 16 mN/m . Additionally, the collapse pressure of pure TsNiPc solution and pure water is 1.4 and 14 mN/m , respectively. In contrast, when TPPS and TsNiPc solutions were used as subphases, the composite membranes exhibited significantly enhanced stability: the CCA/TPPS composite films reached a collapse pressure of 65 mN/m , while the CCA/TsNiPc composite films had a collapse pressure of 58 mN/m . This significant enhancement of the surface pressures is mainly attributed to the multiplicity of intermolecular interactions between the CCA molecules and the dye molecules that arise from interactions between the CCA and dye molecules a high π - π stacking interaction is formed by the massive π -conjugation system in the TPPS, TsNiPc, and CCA molecules. On the other hand, the intermolecular electrostatic attraction also promotes the development of the composite film. The composite film's molecular organization structure becomes more stable and denser. Owing to these excellent properties, the composite film system was successfully created and utilized for further testing.

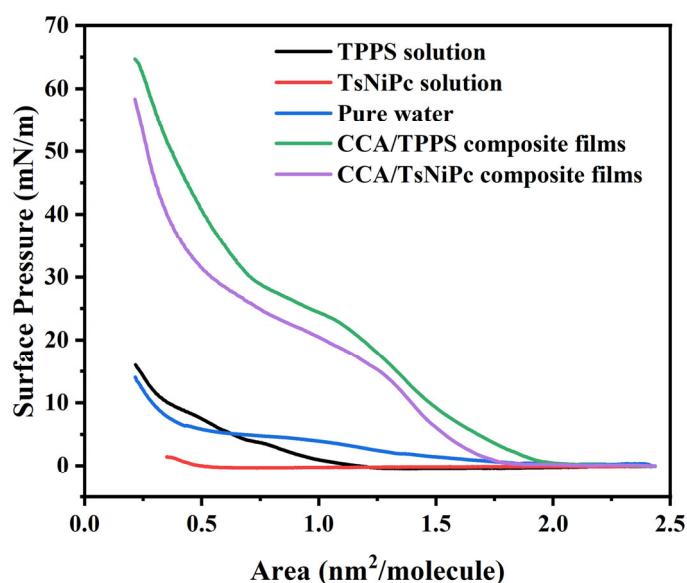


Figure 1. Surface pressure-area isotherm (π -A) plots of $1 \times 10^{-4} \text{ mol/L}$ TPPS solution, $1 \times 10^{-4} \text{ mol/L}$ TsNiPc solution, 0.1 mg/mL CCA solutions on pure water, $1 \times 10^{-4} \text{ mol/L}$ TPPS, $1 \times 10^{-4} \text{ mol/L}$ TsNiPc subphase surfaces.

Atomic force microscopy (AFM) is an important tool for characterizing the surface topography of solid materials. It provides nanoscale resolution of surface topography, including film height and aggregate distribution. A single layer of LB film was placed onto the peeled mica sheet for testing using the vertical lifting technique, and before starting the test, select Tapping Mode, set the image scan rate to 512, and the scanning range to $10.0 \times 10.0 \mu\text{m}$. The figures show that the CCA/TPPS and CCA/TsNiPc composite films had film heights of 2.5 and 8 nm (Figure 2a,b), and widths of both sides of 320 and 190 nm (Figure 2a',b'). R_a is a critical factor in measuring the surface roughness of thin films. R_a for CCA/TPPS and CCA/TsNiPc are 22.1 nm, 0.8 nm. Additionally, the aggregates on the surfaces of the CCA/TPPS composite films were more evenly distributed and structurally denser than those on the other films. π - π stacking, electrostatic attraction, and other intermolecular forces resulted in the interaction of CCA molecules and TPPS and TsNiPc to form a uniform and dense composite film, but due to the different conjugation system and electronic properties of TPPS and TsNiPc, which lead to differences in the strength and mode of their interactions with CCA molecules, this ultimately manifested in the significant differences in the surface morphology of the film.

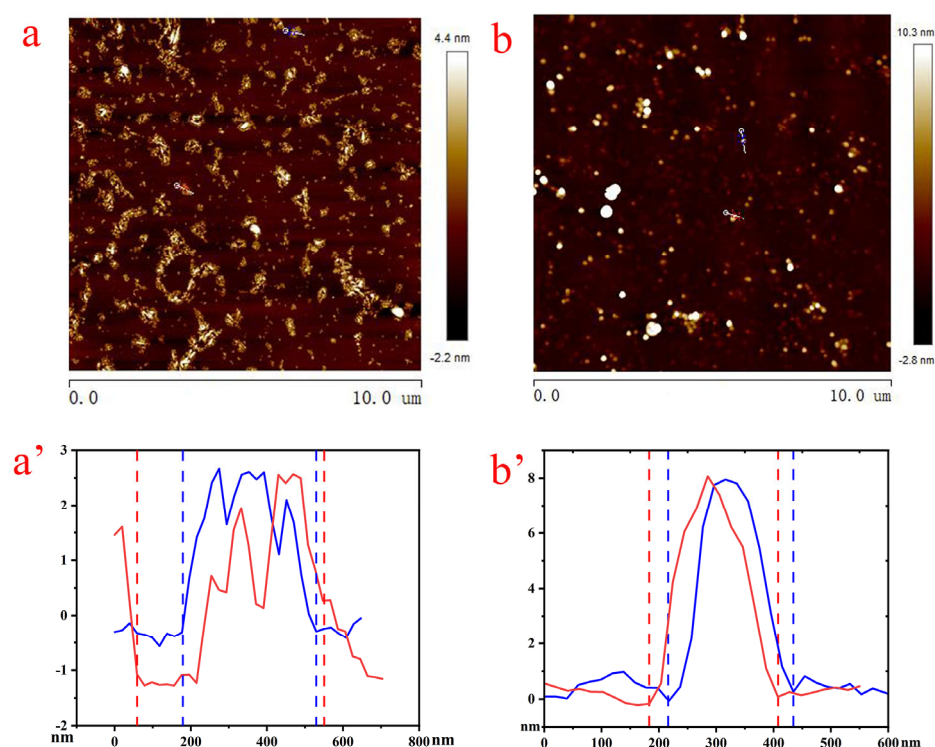


Figure 2. AFM images of (a) CCA/TPPS monolayer film, (b) CCA/TsNiPc monolayer film. Plot of surface height values of (a') CCA/TPPS monolayer film, (b') CCA/TsNiPc monolayer film. The blue and red lines represent two different sampling locations.

Transmission electron microscopy (TEM) was used to acquire high-resolution pictures of the films. Monolayer LB films were transferred to carbon support films using the horizontal transfer method for testing. Figure 3 presents the test results. A consistent flower-like structure was displayed by the CCA/TPPS composite film (Figure 3a), and the flocculent structure was displayed by the CCA/TsNiPc composite film (Figure 3b). CCA combined with both subphases to generate dense, homogeneous composite films. Different molecular aggregation patterns were caused by varying intermolecular force magnitudes and orientations, which produced composite films with various morphologies.

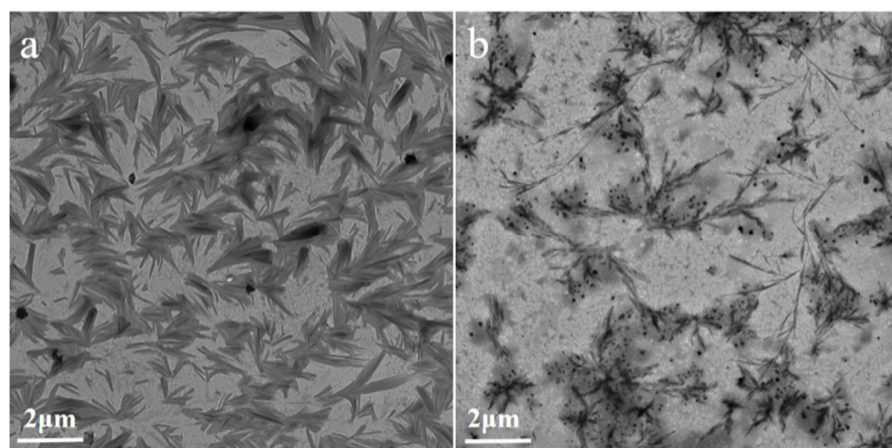


Figure 3. TEM images of (a) CCA/TPPS monolayer film, (b) CCA/TsNiPc monolayer film.

UV-Vis spectroscopy was used to investigate the aggregation behavior between CCA molecules with TPPS and TsNiPc subphases. The wavelength range of the selected UV-absorbing light is 200–800 nm. 60-layer films formed by mixing CCA molecules with the two dyes were transferred onto quartz sheet substrates using the horizontal transfer method for testing. When CCA was added, the high absorption peak at 434 nm in the pure TPPS solution redshifted to 492 nm, and J-aggregation took place (Figure 4a). J-aggregation happens as a result of the interaction between the polar group of the CCA molecule and the sulfonic acid group of the TPPS molecule, which breaks the electronic symmetry of the conjugated system and widens the energy gap. This interaction is the cause of the absorption peak's noticeable blue shift. When TsNiPc bound to the CCA, its absorption peak blueshifted from 624 to 613 nm, resulting in H-aggregation to occur as well (Figure 4b). The absorption peak's blue shift was smaller due to the metal ligand Ni^{2+} in TsNiPc dispersing the intermolecular interactions. To verify the uniformity of the layer thickness, we also performed UV-visible testing of CCA/TPPS composite films with different numbers of layers, and the results showed that the peaks are not shifted with the increase in the number of layers, while the intensity of the absorption values increases consequently. Thus, the uniformity of the thickness of the multilayer composite film is confirmed.

Contact angle tests were performed at room temperature and pressure to realize the hydrophilic and hydrophobic properties of the composite film, with droplet volume of 5 μL per test. The figure shows that the CCA/TPPS and CCA/TsNiPc composite films had contact angles of 92.6° and 101.8° , respectively (Figure 5a,b). Although both composite films demonstrated hydrophobicity, the CCA/TPPS composite film tends to reduce the surface contact angle and exhibits weak hydrophobicity due to the influence of the hydrophilic group, the sulfonic acid group of TPPS. Ni^{2+} forms a stable coordination structure with the surrounding ligands, which enhances the intermolecular close-packing and reduces the exposure of hydrophilic groups, and thus the CCA/TsNiPc composite film exhibits strong hydrophobicity.

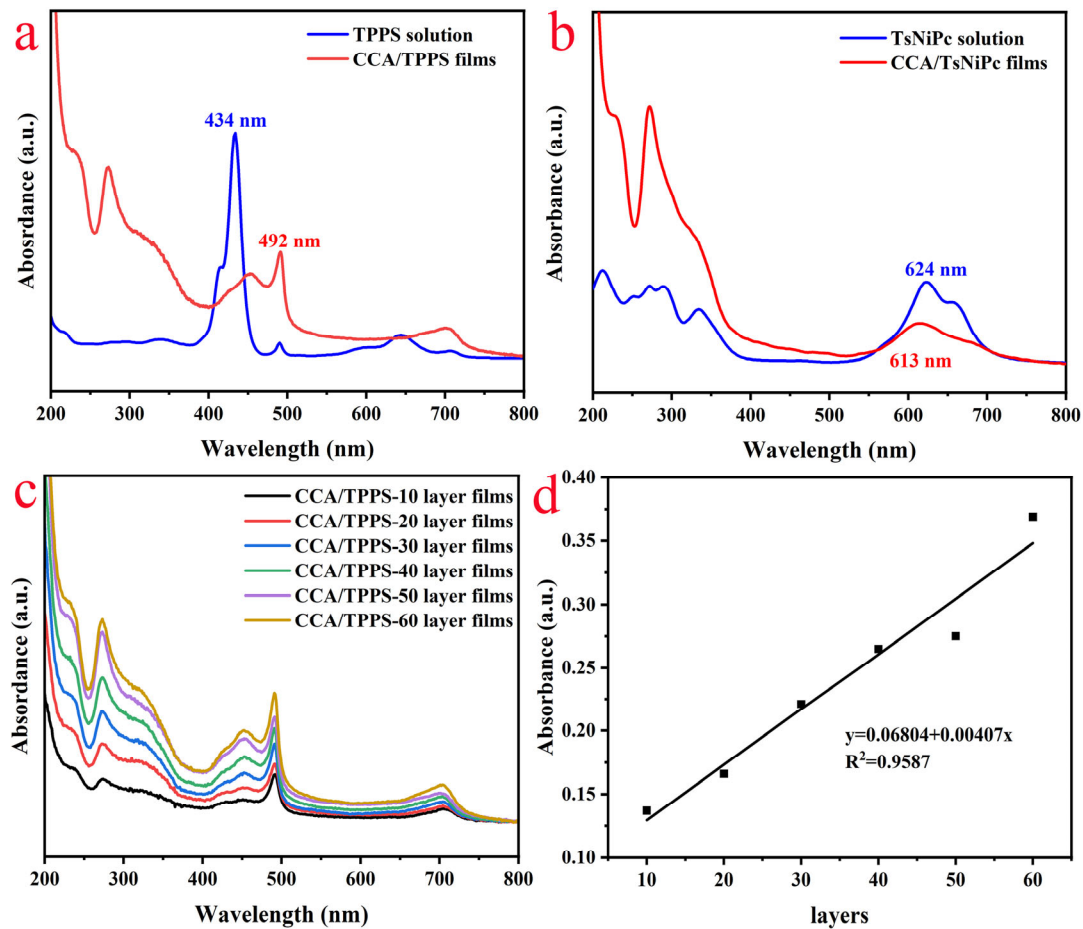


Figure 4. UV-vis spectra of (a) CCA/TPPS-60 layer composite films, (b) CCA/TsNiPc-60 layer composite films, (c) CCA/TPPS composite films with different numbers of layers. (d) Linear fit plot of number of layers versus absorbance.

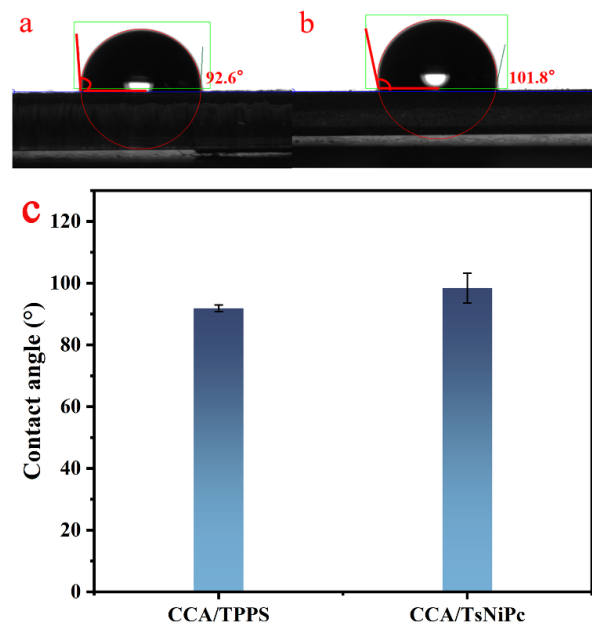


Figure 5. Contact angle images of (a) CCA/TPPS-60 layer composite films, (b) CCA/TsNiPc-60 layer composite films, (c) histogram with error bars.

Chirality is a widespread property in organic molecules, and CCA possesses supramolecular chirality. To investigate the chirality of CCA/TPPS and CCA/TsNiPc composite LB films, the chirality of the two composite films was analyzed using circular dichroism spectroscopy. Figure 6 displays the test findings. In comparison, the CCA/TPPS composite LB film exhibited a strong CD signal at 300–400 nm, while the CD signal of the CCA/TsNiPc composite LB film was almost zero. In an asymmetric environment, the chiral microenvironment created by the CCA and other chiral properties in the surrounding space of the composite film formation with CCA causes the TPPS molecules to react to the chiral-induced circular dichroism by the electron cloud distortion of the TPPS molecules' porphyrin ring. However, because the phthalocyanine ring's rigid planar structure was insensitive to the surrounding chirality, the CCA/TsNiPc composite film failed to show obvious circular dichroism during film formation. As a result, the chirality of CCA had no significant impact on intermolecular stacking and arrangement. Chiral materials are crucial for optical devices, asymmetric catalysis, and molecular recognition, which will also expand the range of applications for the prepared LB films.

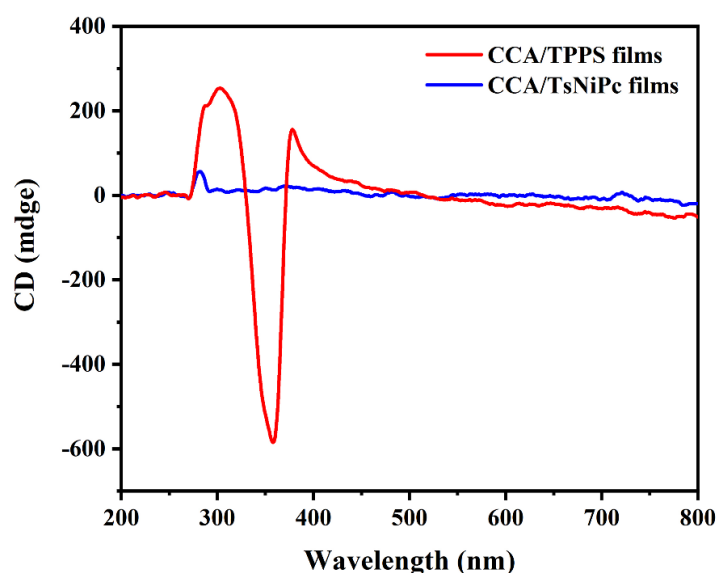


Figure 6. Circular dichroism spectrogram of CCA/TPPS-60 layer and CCA/TsNiPc-60 layer.

SERS uses the adsorption work by adsorbing molecules onto the surface of specific nanostructures, inducing charge transfer between the surface of the composite membrane and R6G molecules, thereby forming electron-hole pairs. When the electron-hole pairs recombine, the molecules resonate and polarize, resulting in a significant enhancement of the Raman signal. TPPS and TsNiPc's massive conjugated planar structures enable charge transfer with CCA. The SERS plots of CCA molecules transported on various subphases of 60-layer composite films are displayed in Figure 7a. Conversely, CCA/TPPS composite films exhibit more prominent SERS augmentation. To gather more detailed information, various layers of CCA/TPPS composite films were chosen for additional testing. The enhancement of the CCA/TPPS-40 composite film was the best among the selected layers (Figure 7b). The ideal SERS substrate should not only have a sensitive response signal, but also have good stability. Therefore, 36 locations' SERS signals were randomly captured on the CCA/TPPS composite film substrate. As shown in Figure 7c, the R6G spectra of 36 random sites are highly consistent, which means that this film has good homogeneity. In addition, Figure 7d,e show that the peak intensity at 614.07 cm^{-1} is highly consistent,

where the relative standard deviation is 12.08%. For the CCA/TPPS-40 composite film, the enhancement factor (EF) can be determined by the following equation:

$$EF = \frac{I_{surf} \times N_{bulk}}{I_{bulk} \times N_{surf}} \quad (1)$$

where N_{bulk} and N_{surf} represent the number of R6G molecules on the film substrate and pure silicon wafer substrate, respectively, and I_{bulk} and I_{surf} are the areas of the Raman vibrational bands in the sample and substrate. After adding 10 μL of a 10^{-3} M R6G solution, the values of I_{surf} and I_{bulk} were obtained at a Raman vibrational band with a center frequency of 614 cm^{-1} . The excitation wavelength chosen for the SERS test was 532 nm. It was found that the I_{surf} value for the CCA/TPPS-40 film was 2.97×10^4 , and the I_{bulk} value was 2.51×10^3 . Therefore, $\frac{I_{surf}}{I_{bulk}}$ has a value of 11.83. For the value of N_{bulk} , it can be calculated using the following equation [43]:

$$N_{bulk} = S_{laser} \times d \times \rho \times \frac{N_A}{M} \quad (2)$$

The laser spot area, S_{laser} , is the area of the irradiated area with a diameter of $1\text{ }\mu\text{m}$. d is the depth of penetration, and the value is $10\text{ }\mu\text{m}$. The density ρ is 0.79 g/cm^3 , the molar mass of the solid R6G is 479.01 g/mol , and N_A is Avogadro's constant of 6.022×10^{23} , which is calculated as $N_{bulk} 0.77 \times 10^{10}$. The area of the R6G molecule is about 2 nm^2 , and the area of laser irradiation is about $0.8\text{ }\mu\text{m}^2$. Therefore, the N_{surf} value can be obtained by calculating the diameters of the two areas to be 4×10^5 .

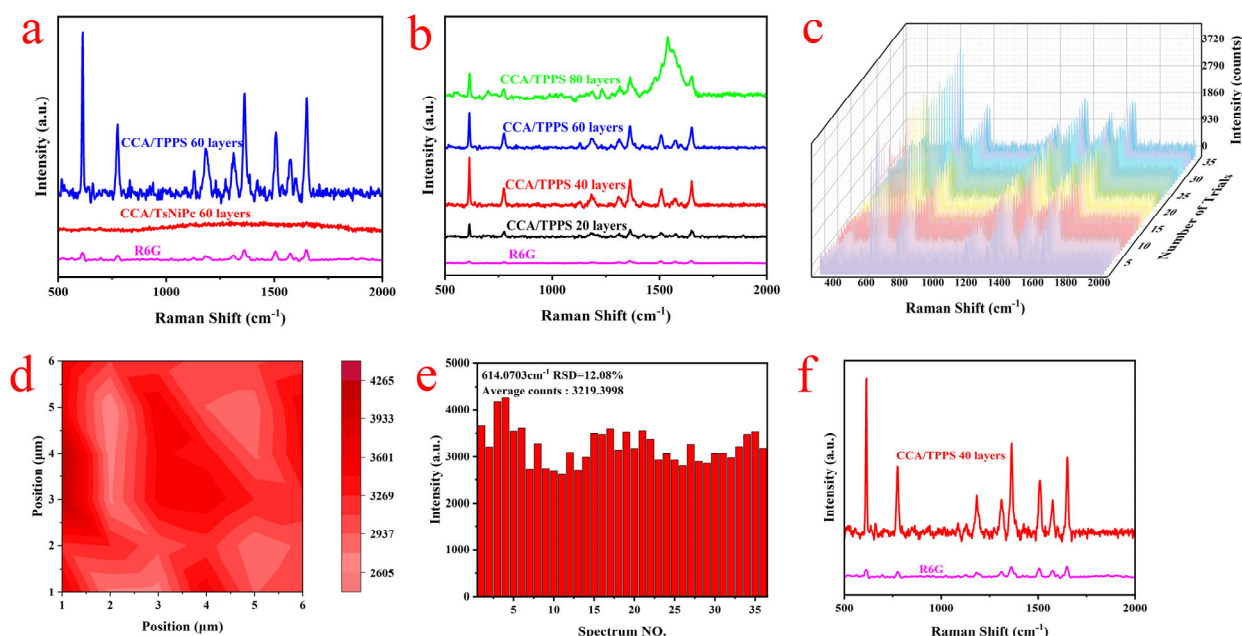


Figure 7. SERS image of R6G molecules (10^{-3} M) on the (a) CCA/TPPS and CCA/TsNiPc composite films of 60 layers, (b) CCA/TPPS composite films with different numbers of layers; (c) The SERS of R6G molecule randomly collected at 36 points on the CCA/TPPS-40 multiple composite films, (d) the point-by-point SERS mapping of R6G molecule on the CCA/TPPS-40 multiple composite films at 614.07 cm^{-1} , (e) The corresponding Raman intensity distribution, (f) The SERS of R6G molecules on the CCA/TPPS-40 multiple composite films substrate.

Substituting the above values into Equation (1), we can calculate the EF of the CCA/TPPS-40 film to be 2.28×10^5 . Low deviation (12.08%) and high EF show that the fabricated composite films are sensitive to the Raman signals, and they have excellent

homogeneity. The computational results demonstrate that the fabricated composite films exhibit responsiveness to Raman signals with excellent homogeneity and stability. Through intermolecular interactions such as hydrogen bonding or π - π stacking, CCA can further improve the charge transfer pathway and change the electron cloud distribution of TPPS. In addition, the large π -conjugated structure of TPPS greatly increases the Raman scattering cross-section of the vibrational modes and enhances the electronic resonance, thus dramatically improving the Raman signal.

5. Conclusions

The LB approach was used in this investigation to prepare CCA/TPPS and-CCA/TsNiPc composite films successfully. To obtain thorough information regarding the morphology and characteristics of the films, a number of experiments were conducted. The films' shape and depth were confirmed by AFM and TEM. Both CCA/TPPS and CCA/TsNiPc composite films exhibited a hydrophobic character. UV-vis demonstrated that both CCAs formed membranes with TPPS and TsNiPc by H aggregation. The electronic transfer of TPPS molecules in a chiral-induced circular dichroism response was caused by the chiral property of CCA. This resulted in a strong CD signal at 300–400 nm for the CCA/TPPS composite film, expanding the potential of using LB films in the field of chiral sensors. In SERS, the CCA/TPPS-40 film's EF is 2.28×10^5 , and its RSD is 12.08%. The great similarity and sensitivity offer a new option for the ideal SERS substrate.

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Conflicts of Interest: The authors declare no conflicts of interest.

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