

# Orientation of 6-Mercaptopurine SAMs at the Silver Electrode as Studied by Raman Mapping and in Situ SERS

Hui Chu,<sup>†</sup> Haifeng Yang,<sup>†,‡</sup> Shuangyan Huan,<sup>†</sup> Guoli Shen,<sup>\*,†</sup> and Ruqin Yu<sup>†</sup>

State Key Laboratory for Chemo/Biosensing and Chemometrics, College of Chemistry and Chemical Engineering, Hunan University, Changsha 410082, People's Republic of China, and Department of Chemistry, Shanghai Normal University, 100 Guilin Road, Shanghai 200234, People's Republic of China

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Self-assembled monolayers (SAMs) of 6-mercaptopurine (6MP) on a silver electrode in acid and alkaline media were investigated by a combination protocol of the SERS technique with Raman mapping, and it was found that the adsorption mode of 6MP SAMs changed with the pH value of the environment. Quantum calculations for the vibrational mode were performed by the BLYP/6-31G method. 6MP was adsorbed on the silver electrode with a tilted orientation via S, N1, and N7 atoms in acid medium, while the SAMs adopted head-on adsorption modes with the S atom and the N1 atom anchoring the silver surface in alkaline medium. However, 6MP SAMs turned to the same upright orientation on the electrode through the S and N7 atoms when either acid or basic solution was removed. Stability of 6MP SAMs was observed by in situ SERS spectroelectrochemical measurements. The results reveal that the desorption potentials of 6MP SAMs formed under acid and alkaline conditions from the Ag electrode were at ca.  $-1.3$  V and  $-1.6$  V vs SCE, respectively.

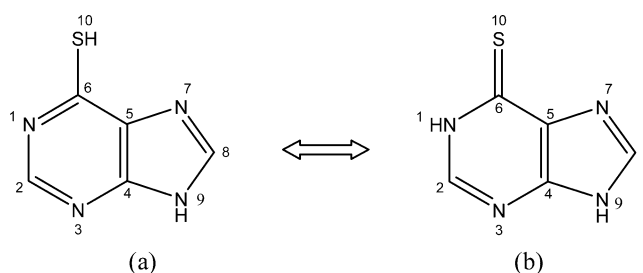
## I. Introduction

6-Mercaptopurine (6MP), one of the oldest chemotherapy drugs which has been utilized for many years, is increasingly used as an active medicine in the treatment of a variety of diseases, including rheumatologic disorder, lymphoblastic leukaemia, inflammatory bowel disease, and prevention of rejection following organ transplantation.<sup>1–5</sup> The coordination chemistry of 6MP has received a great deal of attention because of the potential of linking the drug's coordination to its chemotherapeutic activity.<sup>6</sup> The coordination of 6MP to metals has been studied for producing 6MP–metal complexes acting as slow-release drugs of 6MP<sup>7</sup> and to exploit this reaction for heavy-metal determination.<sup>8,9</sup>

6MP can be modified on various metallic surfaces by the self-assembled monolayers (SAMs) technique, which can offer opportunities to increase our fundamental understanding of self-organization, structure–property relationships, and interfacial phenomena. The investigation of 6MP is of special interest to chemists because of its electronically and stereochemically versatile binding sites. This molecule can attach onto the metallic electrode with possible binding sites of N1, N3, N7, N9, and S10 atoms (See Scheme 1), so there are different adsorption modes of the 6MP molecule at different solid surfaces.

A series of methods including IR,<sup>10</sup> atom force microscopy,<sup>11</sup> conventional electrochemical methods,<sup>11,12</sup> Raman spectroscopy,<sup>14,15</sup> X-ray photoelectron spectroscopy (XPS),<sup>16,17,34</sup> and scanning tunneling microscopy<sup>18</sup> have been employed in studying the structural properties of adsorbed monolayers on solid surfaces. Surface-enhanced Raman spectroscopy (SERS) becomes an especially attractive option because of its high spatial

SCHEME 1: Structure of 6-Mercaptopurine in Thiol Form and Its Tautomer



resolution, ultrasensitivity, and high structural information content.

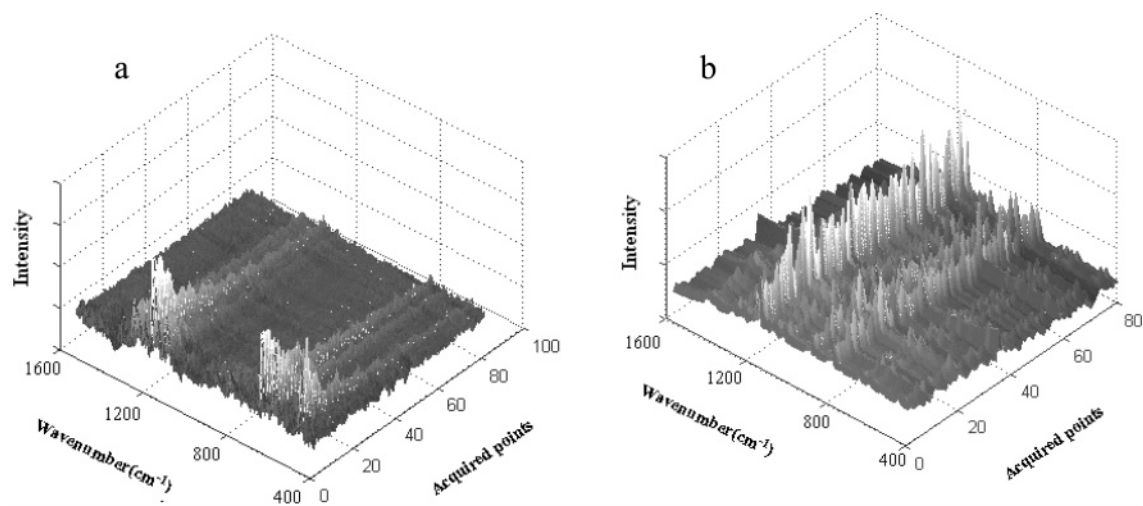
Blázquez et al. have reported the voltammetric studies of 6MP monolayers on Hg and Au surfaces,<sup>19–21</sup> and a cathodic stripping voltammetric study of 6MP at the silver electrode has been carried out by Zeng et al.<sup>22,23</sup> Taniguchi et al.<sup>24</sup> have studied the electrochemical behavior of cytochrome C on a 6MP modified gold electrode and concluded that 6MP would adsorb onto the Au surface through the S10 and N7 in a vertical orientation. SERS spectra of 6MP on gold and silver surfaces have been reported with its orientation in the adsorbed state discussed.<sup>25,26</sup> Hosten and co-workers<sup>25</sup> have employed SERS in determining the orientation of 6MP adsorbed on a silver electrode in basic media, concluding that 6MP attaches head-on through the S10 and N1 atoms to the Ag surfaces.

In this study, we extend the SERS mapping, which has been used for the study of 6MP SAMs on gold surfaces in our previous work,<sup>26</sup> to observe the adsorption procedure of the SAMs at the silver surface in acid and alkaline media. The structural properties of two kinds of 6MP SAMs on silver electrodes were investigated on the basis of the assignment of vibrational modes and SERS mechanism, and their electrochemical stabilities were examined by in situ SERS spectroelectrochemical measurements. In this work, the feature of 6MP

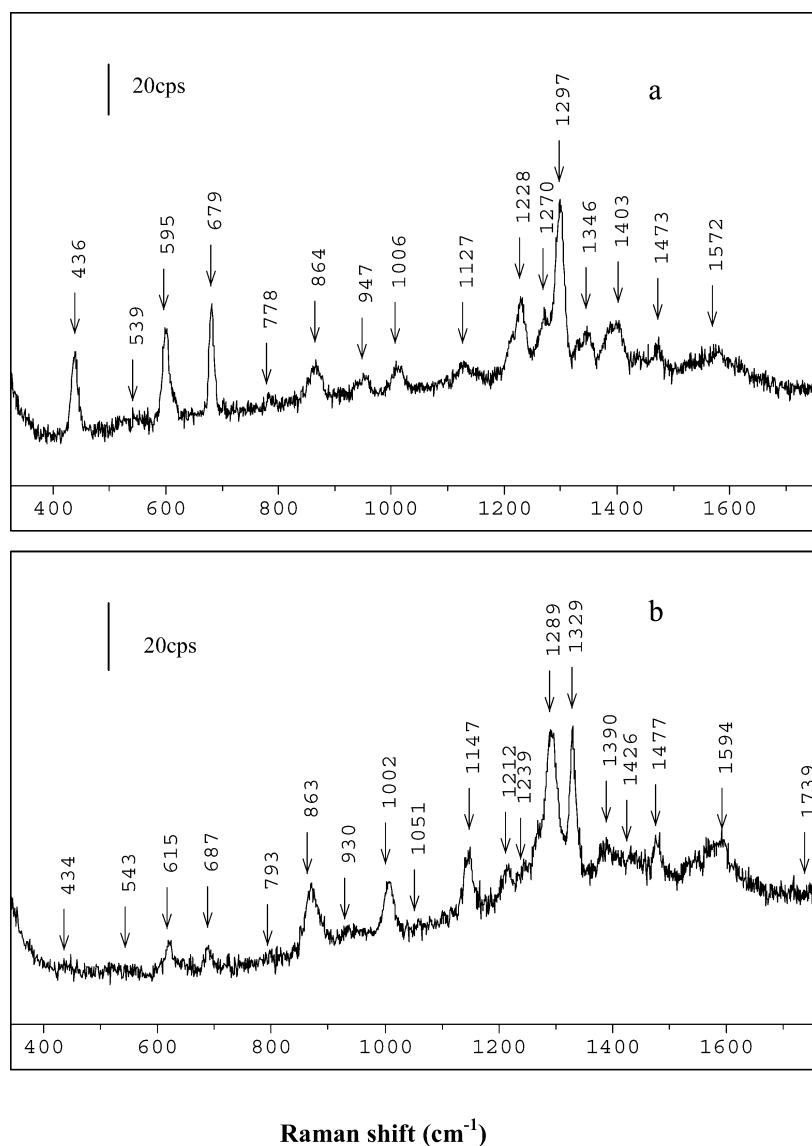
\* Corresponding author. Laboratory for Chemo/Biosensing and Chemometrics, College of Chemistry and Chemical Engineering, Hunan University, Changsha 410082, China. Fax: +86-731-882-2782. Tel: +86-731-882-2237. E-mail: glshen@hnu.cn.

<sup>†</sup> Hunan University.

<sup>‡</sup> Shanghai Normal University.



**Figure 1.** Raman mapping of the formation of SAMs at the roughed silver electrode recorded in  $10^{-4}$  mol/L 6MP in acid (a) and alkaline (b) solutions. The time to take each spectrum is about 50 s.



**Figure 2.** SERS spectrum of 6MP SAMs adsorbed on the Ag electrode recorded in an acid solution ( $\text{pH} = 1.28$ ) (a) and when the acid solution was removed (b).

SAMs on a Ag electrode formed in an acid solution is reported for the first time, and a discussion about the cause of the

different behavior of 6MP SAMs on Ag and Au surfaces is also provided to further our studies on 6MP SAMs.

TABLE 1: Assignments for the SERS Bands of 6MP SAMs at the Roughed Ag Electrode in the Acid State<sup>a</sup>

observed band in an acid solution	SERS bands in solution	SERS bands no solution	calc. freq.	vibrational assignments
441	436s	434vw	391	$\sigma\text{C6-S} + \text{br}(\text{pyrim})$
	538vw	543vw	558	$\nu\text{C2-H} + \nu\text{C8-H} + \nu\text{C2-N3-C4} + \nu\text{C6-S} + \nu\text{C6-C5} + \nu\text{N9-C8}$
588	595s	615m	588	$\text{br}(\text{pyrim}) + \delta\text{C6-S}$
675	679s	687w	677	$\text{br}(\text{pyrim} + \text{imid}) + \nu\text{C8-H} + \nu\text{N9-H}$
	778vw	793vw	825	$\nu\text{C8-H} + \nu\text{C6-C5-C4} + \nu\text{N3-C4}$
	864m	863s	884	$\nu\text{C8-H} + \nu\text{N7-C8} + \nu\text{N9-C8}$
	947m	930vw	945	$\sigma\text{C5-N7-C8} + \delta\text{N9-C8} + \sigma\text{C6-S}$
			969	$\nu\text{C2-H}$
	1006m	1002s		
1023			1018	$\sigma\text{S-H}$
	1127w	1147s	1163	$\text{br}(\text{pyrim} + \text{imid}) + \sigma\text{C6-S} + \delta\text{C2-H} + \delta\text{C8-H}$
		1212sh		
	1228s	1239sh	1238	$\sigma\text{C6-N1-C2} + \delta\text{C8-H} + \sigma\text{C6-S} + \delta\text{C2-N3} + \delta\text{C2-H}$
1265	1270sh		1271	$\sigma\text{C6-N1-C2} + \sigma\text{N7-C8} + \delta\text{C8-H} + \delta\text{C2-H} + \delta\text{N9-H}$
	1297vs	1289vs	1293	$\delta\text{C2-H} + \delta\text{C8-H} + \sigma\text{N1-C2-N3} + \delta\text{N9-H}$
1330	1346m	1329vs	1344	$\sigma\text{C5-N7-C8} + \text{br}(\text{pyrim}) + \nu\text{C8-H} + \nu\text{C2-Hp}$
1377	1403m	1390m	1404	$\delta\text{C2-H} + \text{br}(\text{pyrim} + \text{imid}) + \delta\text{N9-H} + \delta\text{C6-S}$
1421		1426w	1422	$\delta\text{N9-H} + \delta\text{C2-H} + \delta\text{C8-H} + \sigma\text{N9-C8}$
1449	1473w	1477w	1460	$\sigma\text{N1-C2} + \delta\text{C2-H}$
1519			1494	$\delta\text{N7-C8} + \sigma\text{C8-H} + \delta\text{N9-H}$
	1572w	1594m	1588	$\text{br}(\text{pyrim}) + \delta\text{C2-H} + \delta\text{N9-H} + \delta\text{C6-S} + \delta\text{N7-C5} + \delta\text{N9-C4}$

<sup>a</sup> Wavenumbers are given in  $\text{cm}^{-1}$ . Key:  $\delta$ , in-plane vibration;  $\nu$ , out-of-plane bending vibration;  $\sigma$ , stretching vibration; br, ring breathing vibration; tw, twisting vibration; wag, wagging vibration; pyrim, pyrimidine; imid, imidazole; sh, shoulder peak; vs, very strong; s, strong; m, middle; w, weak; vw, very weak.

## II. Experimental Section

**Chemicals.** 6-Mercaptopurine monohydrate (99.5+%) was obtained from Acros Organics and used as received. Two stock solutions of 0.1 M 6MP were prepared by dissolving the solid in 0.01 mol/L HCl and 0.01 mol/L KOH solutions, respectively. Double-distilled water was used throughout to prepare solutions, and pure nitrogen was used for deaerating the working solutions. All other reagents were of analytical grade.

**Roughening the Surface of the Silver Electrode.** The silver electrode was made from polycrystalline silver rods (99.99%). This silver electrode was first mechanically polished with 1.0, 0.3, and 0.05 mm alumina powder to a mirror finish, followed by ultrasonic cleaning with purified water for about 45 s. Then, it was treated in a 0.1 mol/L KCl solution by an oxidation–reduction cycling (ORC) method. After being subjected to the roughening treatment, the electrode was rinsed thoroughly with water and transferred into a homemade cell with a sealed quartz window for Raman measurement. All the electrochemical experiments were conducted by a CHI 760B electrochemical workstation (Shanghai Chenhua instrument, Shanghai, China). The counter electrode was a platinum ring (99.9%). All the potentials were quoted versus a standard calomel electrode (SCE).

**SAMs.** After the cell was installed onto the XY stage below the sampling objective of the Raman spectroscopy, a solution of 6MP with a concentration of  $10^{-4}$  mol/L diluted freshly from the stock solution was injected into the cell to form monolayers at the roughened silver surface. The self-assembling procedure was monitored by Raman mapping measurement.

**Raman Mapping and in Situ SERS Spectroelectrochemical Measurement.** Raman measurements summarized in the previous reports<sup>26</sup> use a Jobin Yvon Micro-Raman spectroscopy (RamLab-010) equipped with an integral Olympus BX40 microscope with a 50 $\times$  objective (8 mm), a holographic grating (1800 g/mm), a notch filter to cut the rayleigh line, and a semiconductor-cooled 1024  $\times$  256 pixels charge-coupled device detector. A laser of 632.8 nm with a power of ca. 5 mW was used as the excitation source for the SERS experiment. Each

spectrum was obtained using two accumulations, and the acquisition time was typically 10–15 s. The mapping area was about  $70 \times 40 \mu\text{m}^2$ , and it was squared randomly on a monochrome image of the surface obtained by an optical axis conjugate camera and a white light source. The Raman band of a silicon wafer at  $519 \text{ cm}^{-1}$  was used to calibrate the spectrometer.

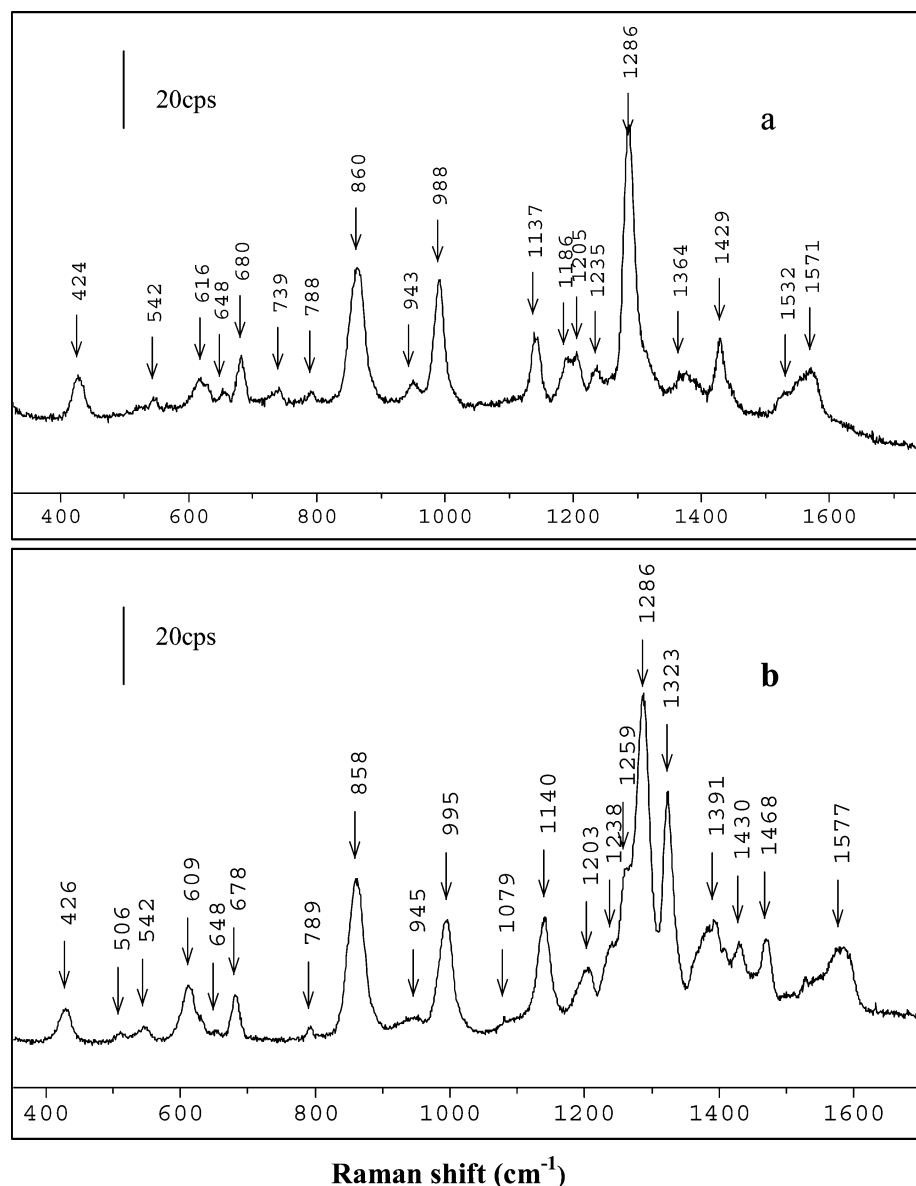
**Theoretical Calculation.** The SERS spectral data of 6MP SAMs were calculated with *Gaussian98* software on the basis of the ab initio BLY/6-31G method. The geometry optimizations of the 6MP–Ag/Au complex were carried out at the B3LYP/LANL2DZ level.

## III. Results and Discussions

**Raman Mapping of 6MP SAMs at the Silver Electrode in Acid and Basic Media.** Raman mapping provides a particularly useful tool for obtaining an overview of the spontaneous SAMs adsorption process.<sup>27</sup> Here, a Raman mapping experiment was conducted to examine the adsorption fashion of 6MP SAMs on the roughened silver surfaces.

Recently, Willey et al.<sup>34</sup> reported that alkanethiols or other organics adsorbed on gold through gold–thiolate bonding degrade rapidly when exposed to air by using XPS and NAXAFS. But in our experiment, the power of the excitation laser was so low that it did not affect the orientation of the 6MP SAMs on the silver surface. The SERS signals could hold the line after 12 h.

Figure 1a,b presents the Raman mapping spectra of 6MP SAMs on the silver surface under acid and alkaline conditions recorded after a queue time of 30 min. The recorded profiles of the Raman spectra obtained at various points on the surface are largely ordered and uniform, and there is almost no change of Raman mapping spectra after a queue time of 12 h (which is not given here). In comparison to 6MP SAMs formed on the gold electrode which need a dynamically self-organized procedure,<sup>26</sup> it could be concluded that formations of 6MP SAMs on the silver electrode in acidic or basic medium are quite quick processes.



**Figure 3.** SERS spectra of 6MP SAMs adsorbed on Ag electrode recorded in an alkaline solution (pH = 12.8) (a) and when the alkaline solution was removed (b).

Figure 2a shows the SERS spectrum of 6MP SAMs adsorbed on a silver electrode in a solution with pH of 1.82, and Figure 2b shows the SERS spectrum recorded after the working acid solution was removed. Table 1 contains these observed SERS bands of 6MP SAMs in acid state along with the relevant assignments based on the quantum calculation results for vibrational modes. It can be found that the band of the S–H vibration at  $1023\text{ cm}^{-1}$  is unobservable and the band at  $441\text{ cm}^{-1}$  corresponding to the C6–S stretching mode in the solution spectrum is shifted to a lower wavenumber at  $436\text{ cm}^{-1}$ . These observations suggest that 6MP should adsorb on the Ag surfaces through the S atom upon the cleavage of the S–H bond, which also leads to the occurrence of the bands at  $1127$  and  $1572\text{ cm}^{-1}$  resulting from the deformation vibration of the C2 atom of the pyrimidine ring along with the breathing vibration of this ring. The new band of  $1006\text{ cm}^{-1}$  is related to the Ag–S vibration, which is close to the strong Ag–S stretching frequency at  $1005\text{ cm}^{-1}$  observed by Hatta et al.<sup>33</sup> The strongest band arises at  $1297\text{ cm}^{-1}$  because of the C2–H and C8–H deformation vibrations and the N1–C2–N3 stretching vibration, while the band at  $595\text{ cm}^{-1}$  assigned to the breathing vibration of the

pyrimidine ring and the band at  $679\text{ cm}^{-1}$  attributed to the breathing modes of the pyrimidine and imidazole rings also possess strong intensities. The breathing vibration of the pyrimidine moiety and the deformation mode of the C8–H and C2–H co-contributing to the band at  $1346\text{ cm}^{-1}$  become clear. Therefore, a conclusion that 6MP might adopt a tilted orientation on the silver surfaces under acidic condition via the Ag–S bond could be drawn, and the N1 and N7 atoms might both contribute to this kind of attachment.

From Figure 2b, the SERS spectrum exhibits an obvious change in comparison with the SERS spectrum of Figure 2a. The band at  $863\text{ cm}^{-1}$  mostly attributable to the C8–H out-of-plane bending vibration increases, while deformation vibrations of the N9–H, C2–H, and C8–H found at  $1421\text{ cm}^{-1}$  in the solution Raman spectrum and unobserved in the spectra of Figure 2a occur at  $1426\text{ cm}^{-1}$  in the spectrum in Figure 2b. Moreover, the band at  $1329\text{ cm}^{-1}$  resulting from the C8–H and C2–H deformation vibrations along with the breathing vibration of the pyrimidine ring shows remarkable enhancement. These observations indicate that the 6MP molecules assume the standing-up mode on the silver surfaces. This conclusion is

**TABLE 2: Assignments for the SERS Bands of 6MP SAMs at the Roughened Ag Electrode in the Basic State<sup>a</sup>**

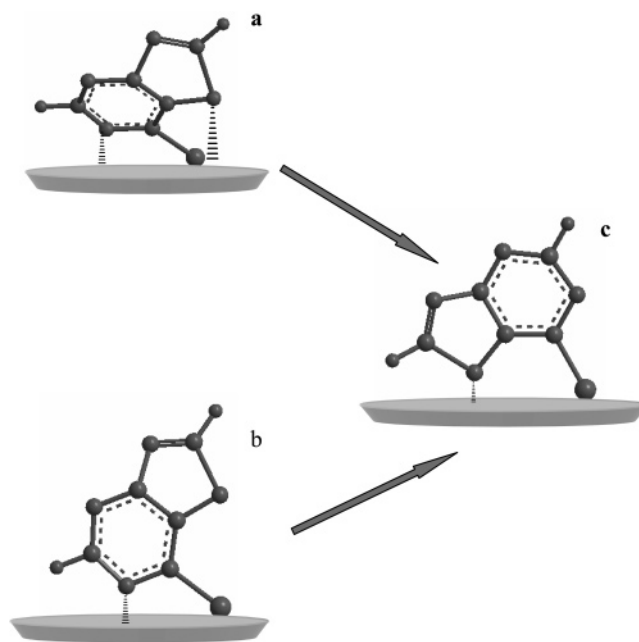
observed bands in an alkaline solution	SERS bands	calc. freq. tautomer	calc. freq. thiol form	vibrational assignments
440	424m	430		br(pyrim)+ $\sigma$ C6-S
	542w	566		$\nu$ C2-H+ $\nu$ C2-N3 + $\nu$ N1-C2+ $\nu$ C8-N9+ $\nu$ C6-S
605	616m	606		$\delta$ C6-S+ $\delta$ N1-H+wag(pyrim)+ $\delta$ C2-H+ $\delta$ C8-H+ $\delta$ N9-H
	648w		645	$\nu$ N9-H+ $\sigma$ N1-C2+ $\sigma$ C6-C5-C4
685	680m		677	br(pyrim)+ $\delta$ C6-S
	739vw			
	788w	797		tw(pyrim+imid)+ $\delta$ C2-H+ $\delta$ C8-H+ $\delta$ N1-H
876	860s	860		$\nu$ C8-H
	943w	950		$\nu$ C2-H+ $\nu$ C6-N1-C2+ $\nu$ N1-H+br(imid)+ $\sigma$ C6-S+ $\delta$ N1-C6
1006	988s			
1136	1137s		1162	$\delta$ S-H+ $\sigma$ N1-C2-N3+ $\delta$ C6-S+ $\delta$ C8-H
	1186m	1188		br(pyrim)+ $\sigma$ C6-S + $\delta$ N1-H
	1205sh			
1244	1235w	1236		$\nu$ C8-H+ $\sigma$ C6-S+ $\sigma$ N7-C5+ $\sigma$ N1-C2+ $\nu$ N9-H
1299	1286vs		1293	$\sigma$ N1-C2-C3+ $\delta$ N9-H+ $\delta$ C8-H
1371	1364w	1368		$\delta$ N1-H+ $\nu$ C2-H+ $\nu$ N9-H+ $\nu$ C8-H+br(imid)+ $\sigma$ C6-C5
1410				
1432	1429m	1438		$\nu$ C2-H+ $\nu$ N1-H+ $\nu$ N9-H+ $\sigma$ N1C2+ $\nu$ C4-C5+ $\sigma$ N7-C8
	1532sh			
	1571m	1583		$\delta$ C8-H+ $\sigma$ C8-N7+ $\delta$ N1-H+ $\sigma$ C5-C4-N3+ $\delta$ N9-C4
1596				

<sup>a</sup> Wavenumbers are given in  $\text{cm}^{-1}$ . Key:  $\delta$ , in-plane vibration;  $\nu$ , out-of-plane bending vibration;  $\sigma$ , stretching vibration; br, ring breathing vibration; tw, twisting vibration; wag, wagging vibration; pyrim, pyrimidine; imid, imidazole; sh, shoulder peak; vs, very strong; s, strong; m, middle; w, weak; vw, very weak.

drawn from a simple rule for determining the surface geometry of a planar molecule adsorbed on silver surface, which is based on the observation of a strong or weak aromatic C-H stretching band in a SERS spectrum. As the molecule stands up on the surface, the C-H bond would experience some enhancement.<sup>28</sup> The more obvious band at  $1594\text{ cm}^{-1}$  due to the C2-H and N9-H rocking vibration confirms this conclusion. The intensity of the band at  $687\text{ cm}^{-1}$  attributed to the breathing fashions of the pyrimidine and imidazole rings falls off dramatically, which also suggests that the angle between the rings and the Ag surface becomes large and the molecule might stand up on the surfaces. It can be seen that the bands at  $947$  and  $1345\text{ cm}^{-1}$  related to the stretching vibration of the C5-N7-C8 in the SERS spectrum recorded in the acid solution shift downward to  $930$  and  $1329\text{ cm}^{-1}$  in the SERS spectrum recorded with the acid solution removed. The band at  $1227\text{ cm}^{-1}$  associated with the deformation vibration of the C6-N1-C2 upshifts about  $12\text{ cm}^{-1}$ . It shows that the N7 atom of the imidazole moiety should be located closer to the SERS-active Ag surfaces, but the N1 atom of the pyrimidine moiety might turn away from the surfaces. 6MP might stand up on the Ag electrode via merely S and N7 atoms under this situation.

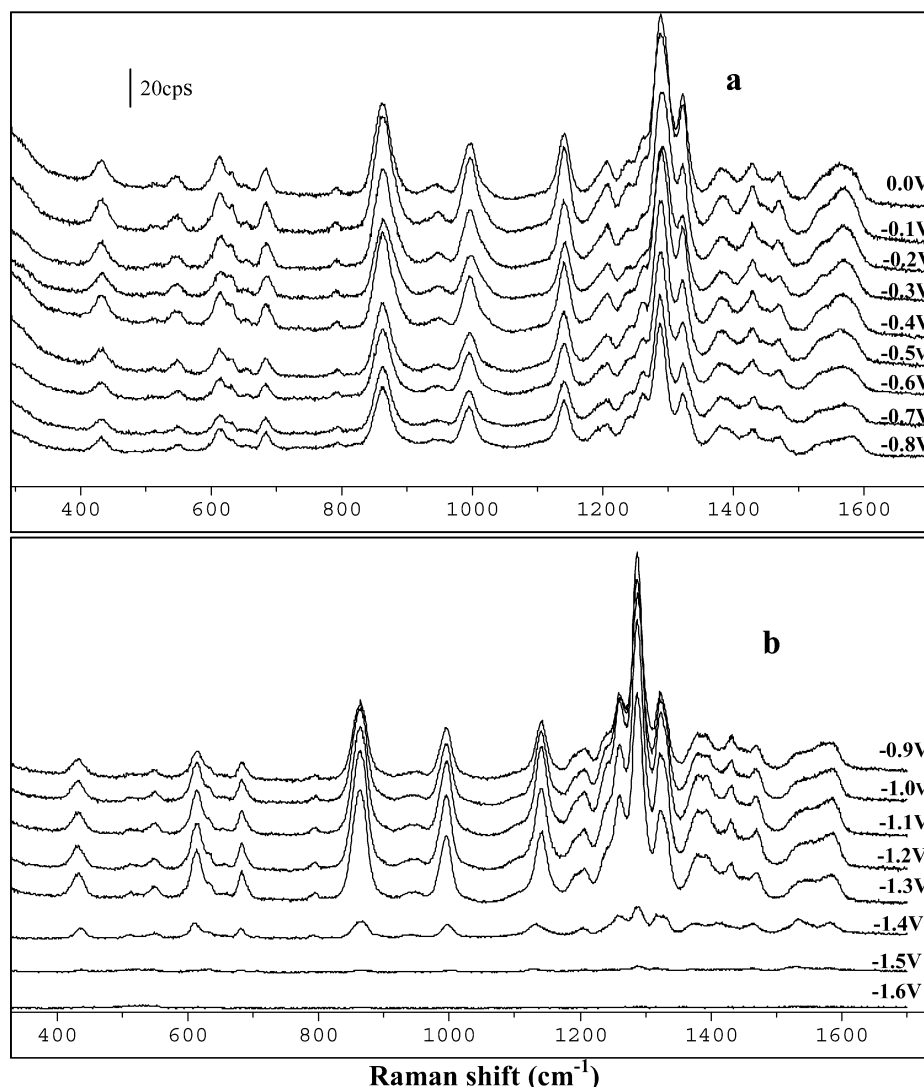
As shown in Figure 3a, the SERS spectrum of 6MP on the silver electrode in a solution with pH of 12.8 was recorded. The corresponding assignments for the observed SERS bands of 6MP SAMs in this state are listed in Table 2. It is observed that the SERS spectrum of 6MP on the silver electrode under basic condition obviously differs from that under acidic condition, and certainly, the adsorptions of 6MP onto the Ag surfaces in acidic and basic states are different. We have found out that 6MP SAMs adopted the same adsorption mode through the S atom of the pyrimidine ring and the N7 atom of the imidazole moiety, anchoring the gold surface in a vertical way in acidic and alkaline media. However, the SERS spectra of 6MP on the roughened silver electrode in alkaline media with a 488-nm laser excitation have been studied by Hosten et al., and they suggest that 6MP attaches head-on through the S and N1 atoms to a silver electrode surface.

The band at  $1186\text{ cm}^{-1}$  resulting from the contribution of the breathing vibration of the pyrimidine moiety and the

**SCHEME 2: The Suggested Adsorption Modes of 6MP SAMs at the Ag Surface (a) in an Acid Solution, (b) in an Alkaline Solution, and (c) after the Acid or Alkaline Solution Was Removed**

stretching vibration of the C6-S appears, while the bands near  $440$  and  $685\text{ cm}^{-1}$  in the alkaline solution spectrum assigned to the breathing mode of the pyrimidine ring and the deformation vibration of the C6-S are downshifted about  $16$  and  $5\text{ cm}^{-1}$ , respectively. These data depict that 6MP attaches head-on to the silver surface through the S atom. The band at  $1299\text{ cm}^{-1}$  of the Raman spectrum in the solution associated with the stretching vibration of N1-C2-N3 is shifted to the lower wavenumber. It might be the attachment of N1 to the Ag atom, which is confirmed by the occurrence of the bands of  $542$ ,  $648$ , and  $943\text{ cm}^{-1}$  due to the vibrations involving the N1 atom. From the above discussion, one concludes that 6MP could adsorb onto the silver electrode through the S atom and the N1 atom, in





**Figure 4.** In situ SERS spectra of the 6MP SAMs formed under alkaline condition on the Ag surfaces recorded in a 0.1 mol/L KCl solution with shift of the applied voltages vs SCE. (a) 0.0 ~ -0.8 V; (b) -0.9 ~ -1.6 V.

agreement with the conclusion drawn by Hosten et al. Figure 3b gives the SERS spectrum of 6MP on the Ag electrode when the basic solution was removed. It is interesting to note that this spectrum is very similar to that of Figure 2b, which implies that the resulting 6MP SAMs adopt the same adsorption mode after the acidic or basic solution is removed.

In acid solution, the protonated N7 atom and N1 atom present the similar potential of anchoring the silver surface; in alkaline solution, the hydroxyl might increase the steric hindrance of the action of the N7 atom with the silver surface, and the N1 atom also presents a low potential of attaching onto the Ag surface. Nevertheless, 6MP SAMs adopt the more stable adsorption mode through S and N7 atoms anchoring the Ag electrode as the acidic or basic solution is removed. Hosten et al. have reported that an attachment through S10 and N7 is 5.80 kcal/mol more stable than the one through N1 by calculating the heats of formation of the various forms of attachment with the PM3 method.<sup>25</sup>

The possible modes of 6MP SAMs on the Ag electrode are presented in Scheme 2.

**In Situ SERS Spectroelectrochemical Measurements.** After removing the 6MP solution from the cell and washing the Ag electrode with doubly purified water, a 0.1 mol/L KCl solution was injected into the cell as the supporting electrolyte. In situ SERS spectroelectrochemical techniques were then carried out

to examine the stability of the 6MP SAMs on the Ag electrode under different potentials.

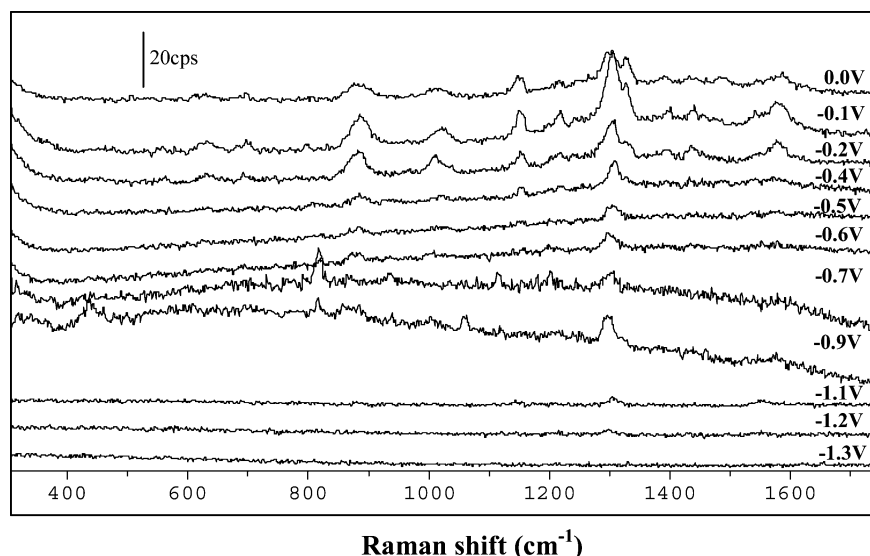
In situ SERS spectroelectrochemical spectra of 6MP SAMs on the silver electrode formed in acidic and basic media are shown in Figures 4 and 5.

When the potential was applied, the bands of the SERS spectra of two kinds of 6MP SAMs increased, indicating that 6MP should be chemically adsorbed onto the silver electrode in acidic and alkaline media.

From Figure 4, the SERS signals decreased abruptly when the potential was shifted from -1.4 V to -1.5 V vs SCE. Then, there was almost no signal as the potential was moved to -1.6 V. The 6MP molecules were entirely detached from the surface at that time.

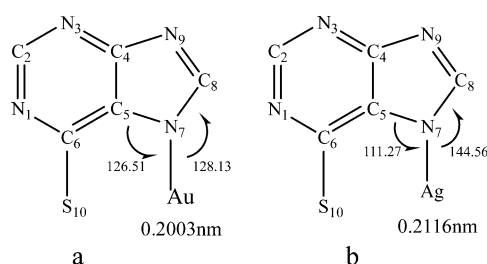
Moreover, the SERS spectra of 6MP SAMs formed under pH of 1.28 change dramatically when the potential was at -0.7 V. It implies that there might be a disorder of 6MP SAMs on the Ag surface due to the partial desorption of the molecules at that potential. The signals are lost at -1.3 V, showing the entire desorption of the 6MP molecules from the Ag electrode.

Zeng and Purdy carried out the cathodic stripping voltammetric studies of 6MP at a silver electrode and found that, when the potential scan range extended sufficiently negative to -1.55 V, the cyclic voltammograms (CVs) of 6MP would acquire a different shape.<sup>22</sup>



**Figure 5.** In situ SERS spectra of the 6MP SAMs formed on the Ag surfaces under acid condition recorded in a 0.1 mol/L KCl solution with shift of the applied voltages from 0.0 to  $-1.3$  V vs SCE.

**SCHEME 3: Plausible Structures of (a) Au-6MP (N7-deprotonated) and (b) Ag-6MP (N7-deprotonated) Complexes at the B3LYP/LANL2DZ Level**



**The Origin of the Different Behavior of 6MP on Silver and Gold Surfaces.** In comparison to the previous work about 6MP SAMs at the gold electrode, the SAMs on the silver substrates in solution exhibit different adsorption modes and different desorption potentials.

Bryant and Pemberton have compared 1-alkanethiols SAMs on Au to those on Ag and suggested that the metal–sulfur bonding strongly influenced the different orientations at Au and Ag.<sup>29</sup> In the case of 6MP SAMs, the metal–nitrogen bonding is also an essential factor in determining the final molecular orientation. As mentioned above, the attachment of 6MP through S10 and N7 is the more stable form, and the SAMs on the Ag and Au surfaces both adopt this form when the solution is removed with no impact of  $H^+$  or  $OH^-$  from the solution. Here, the geometry optimizations for the Au–N7 deprotonated form and the Ag–N7 deprotonated form were carried out at the B3LYP/LANL2DZ level. The plausible structures of Au–6MP and Ag–6MP for the N7 deprotonated forms are drawn in Scheme 3 with their optimized geometry variables. The Au–N7 bond lengths are 0.2003 and 0.0113 nm smaller than Ag–N7 bond length, which indicates that the N7 deprotonated Au complex is more stable than the N7 deprotonated Ag complex. It is also why the pH value of the environment can influence the adsorption mode of 6MP SAMs on Ag easily.

Ulman reported that in SAMs of thiolates on Ag(111) there are 26% more chains per unit area than on Au(111) on-top sites and suggested that the combination of lateral discrimination and electrostatic effects distinguishes the chemisorption of thiolates on Au(111) from that on Ag(111).<sup>30</sup> The sulfur hybridization

is different on the two metals (sp on Ag compared to  $sp^3$  on Au),<sup>31</sup> which leads to a difference in the strength of the substrate–metal interaction and has a profound influence on the behavior of SAMs on the metal. The proposition given by Cho et al. is that the number of atoms on the Au surface is smaller on average than those on the Ag surface.<sup>32</sup> In our work, the entire desorption of 6MP SAMs from the metal surface is finally determined by the sulfur–metal bond. The desorption potential of 6MP on Ag is more negative than that on Au, proving that the sulfur–Ag bond of 6MP is more stable than its sulfur–Au bond.

#### IV. Conclusion

The formation process of 6MP SAMs on the SERS-active Ag electrode has been studied in acidic and alkaline media by the Raman mapping technique, and the corresponding molecular orientation of the adsorbate on the silver surfaces was deduced by attributions of SERS bands on the basis of the results of BLYP/6-31G calculation and the SERS mechanism. The electrochemical stability of the 6MP SAMs on the Ag electrode has been examined by using in situ spectroelectrochemical measurements. Some conclusions could be reached:

(1) The self-assembling of 6MP at the silver surface is a relatively quick process. The 6MP molecule tends to attach onto the Ag electrode in acid media at an angled orientation through the S, N1, and N7 atoms, whereas the sites of interactions with the silver surface under basic condition should be the S atom and the N1 atom of the pyrimidine moiety. However, the adsorption patterns both change into the same modes after the solution is removed, and the corresponding mode is the standing-up orientation via S and N1 atoms.

(2) The adsorption mode of 6MP SAMs obtained at pH of 1.28 changes as the potential is applied to  $-0.7$  V vs SCE, and its desorption potential is around  $-1.3$  V. The SAMs of 6MP formed at pH of 12.8 are detached from the silver surface at the potential of  $-1.6$  V.

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