

Laser Chem. 1988, Vol. 9, pp. 401–416
© 1988 Harwood Academic Publishers GmbH
Photocopying permitted by license only
Reprints available directly from the Publisher
Printed in the United Kingdom

Surface-Enhanced Raman Spectroscopy of Thionine-modified Gold Electrodes

H. R. VIRDEE and R. E. HESTER

Department of Chemistry, University of York, Heslington, York YO1 5DD

(Received 20 May, 1988)

Surface enhanced Raman (SER) and resonance Raman (SERR) techniques have been used *in situ* to investigate thionine-modified gold electrodes. New surface roughening procedures for gold electrodes have resulted in an order of magnitude increase in the Raman signals. As a result of this, Raman spectra from leucothionine have been observed for the first time. The surface Raman spectra of both thionine and leucothionine are essentially unchanged over the pH range from 1.3 to 7 but both show major changes at pH 10. This behaviour has been related to changes in the absorption spectrum of thionine at pH 1.0 where the compound is believed to exist as thionine hydroxide. At pH 1.3 and 7 the Raman signals from thionine arise from a combination of surface enhancement and resonance enhancement processes, whereas signals from leucothionine arise solely from surface enhancement. At pH 10 surface enhancement processes give rise to Raman intensity for both thionine and leucothionine.

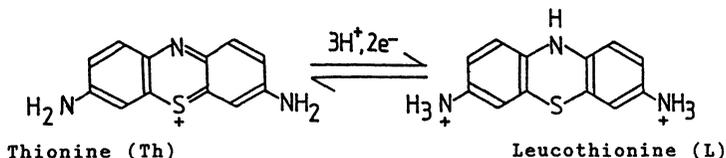
KEY WORDS: Raman spectroscopy, SERS, SERRS, thionine, modified electrodes, resonance Raman.

INTRODUCTION

Electrode surfaces modified by the immobilization of electrochemically active species have attracted considerable research interest in recent years,¹ mainly because of their promise in applications to such processes as electrocatalysis² and solar energy conversion.³ One such

system that has been investigated extensively by both electrochemical⁴⁻⁸ and spectroscopic techniques^{9,10} involves thionine-modified electrodes.

The thionine redox couple in aqueous acid solution may be represented as follows:



Thionine may be electrochemically coated^{4,5} onto substrates such as platinum and gold at oxidizing potentials, thus producing thionine-modified electrodes. The thionine coat formed on the electrode surface is electroactive and can be switched between leucothionine and thionine forms. Electrochemical^{6,7} and x-ray photoelectron spectroscopic studies⁹ have suggested that thionine is dimerized on the electrode surface.

More recently, in this laboratory, surface enhanced Raman spectroscopy (SERS) and surface enhanced resonance Raman spectroscopy (SERRS)¹⁰ have been applied, *in situ*, to the study of thionine-modified gold electrodes in acid electrolyte solution. In that work good quality Raman spectra were observed from the coated thionine. However, no Raman spectra of leucothionine could be observed. The Raman spectrum of coated thionine¹⁰ showed additional bands that were not present in the spectrum of solid thionine. The bands have been attributed to the bridging C—N—C bonds in the thionine dimer formed on the electrode surface.

In this work we present further SERRS and SERS results from thionine and leucothionine on gold electrodes at various pH values.

EXPERIMENTAL

Thionine used in this work was from Fluka and was purified by column chromatography on alumina, washed with chloroform and acetone and dried *in vacuo* at room temperature. Sulphuric acid was laboratory reagent grade from BDH. Sodium hydroxide and potassium chloride

were from Fisons. All the solutions were prepared in distilled water. The electrolyte solutions were deoxygenated by bubbling N_2 for about half an hour prior to the spectroelectrochemical measurements.

The details of the Raman spectroelectrochemical cell, with a rotating electrode, are given elsewhere.¹⁴ An Oxford Electrodes potentiostat was used for the control of potential. A Hewlett-Packard model 70158 X-Y recorder was used for recording the cyclic voltammograms. All the potentials were measured and are reported with respect to a saturated calomel reference electrode (SCE). The gold working electrode used in this work consisted of a *ca.* 7 mm diameter cylindrical disc, *ca.* 5 mm thick, embedded in a Teflon sleeve which was attached to a rotating shaft assembly. The exposed area of the working electrode was 0.385 cm^2 . For all the Raman measurements the working electrode was rotated at *ca.* 3700 r.p.m.

Raman spectra were recorded on a Spex model 1403 spectrometer fitted with a cooled photomultiplier, RCA type C31034-A02, and linked to a Spex DPC-2 digital photometer. A compudrive unit and Scamp minicomputer controlled the instrument. Radiation for exciting the Raman spectra was provided by a Spectra Physics model 170 krypton ion laser. In all experiments the 647.1 nm line was used to excite the Raman spectra and the laser power used was *ca.* 100 mW at the sample. Spectra were recorded with a spectral band pass of 5 cm^{-1} . The scattered light was collected at 90° to the incident laser beam which was itself polarized perpendicular to the scattering plane. The working electrode surface was parallel to the cell window and was set at *ca.* $50\text{--}60^\circ$ with respect to the laser beam.

A roughening procedure for the gold electrode, similar to that described recently by Weaver *et al.*,¹¹⁻¹³ was used prior to the coating of the electrode with thionine. The working gold electrode was polished to a mirror finish with a slurry of alumina polishing powder (Banner Scientific) in distilled water. The electrode was rinsed with distilled water. The Raman spectroelectrochemical cell together with the working electrode was then assembled and fitted with 1 mol dm^{-3} KCl solution in distilled water. After deoxygenating for *ca.* 0.5 hour the oxidation/reduction cycles (ORCs) were then carried out between the potential limits of -0.20 V to $+1.10 \text{ V}$ (SCE) at a sweep rate of 0.05 V s^{-1} . It was found that *ca.* 25 ORCs were necessary for the activation of the gold electrode for optimal surface enhancement.

After the ORCs had been carried out the KCl solution was removed

and the cell was rinsed with distilled water, followed by a rinse with a saturated solution of thionine in $0.05 \text{ mol dm}^{-3} \text{ H}_2\text{SO}_4$. The cell was then filled with thionine solution and the gold electrode was coated⁴ at 1.15 V for 10 minutes. Finally, the thionine solution was removed from the cell and the whole system rinsed with distilled water. Raman spectra of coated thionine were then recorded in $0.05 \text{ mol dm}^{-3} \text{ H}_2\text{SO}_4$ solution at open circuit potential 0.33 V (SCE) using 647.1 nm excitation. The cell was then brought under potential control and the cyclic voltammogram of coated thionine was recorded in the potential range +0.5 V to -0.20 V. A typical cyclic voltammogram from a thionine coated gold electrode in acid solution is shown in Figure 1. Raman spectra of coated thionine were recorded at various applied potentials in the range -0.20 to +0.40 V.

Raman spectra of coated thionine in contact with electrolyte solution at pH 7 and 10 were also recorded. In these cases the pH of the original $0.05 \text{ mol dm}^{-3} \text{ H}_2\text{SO}_4$ solution was changed by adding a concentrated solution of NaOH dropwise until the required pH was reached. A typical cyclic voltammogram of the thionine modified electrode at pH 10 is shown in Figure 1. At pH 7, Raman spectra at

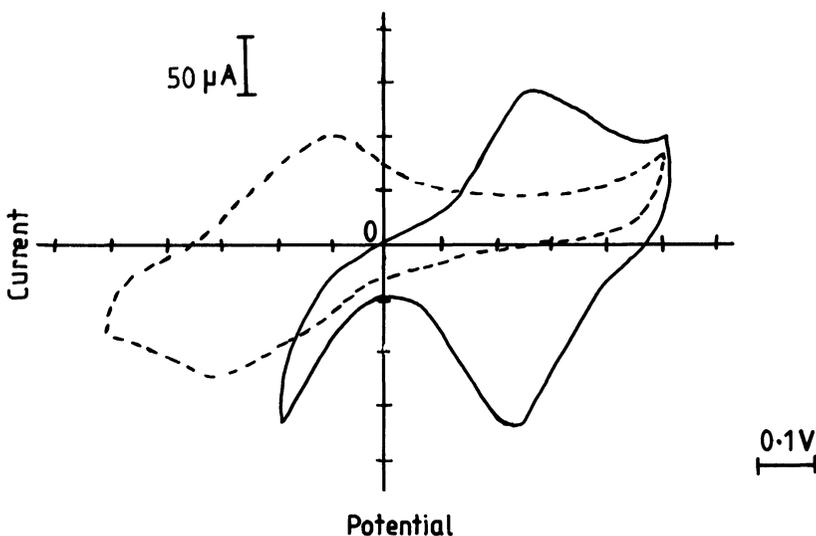


Figure 1 Cyclic voltammogram of thionine-coated gold electrode at pH = 1.3 (solid line) and pH = 10 (broken line); sweep rate 0.05 V s^{-1} .

applied potentials of +0.30 and -0.20 V were recorded. At pH 10, Raman spectra at various applied potentials in the range -0.50 to +0.30 V were recorded.

RESULTS AND DISCUSSION

Raman spectra of thionine-coated gold electrodes in contact with electrolyte solutions at pH 1.3, 7 and 10 have been measured at various applied potentials. For these measurements the gold electrode was activated for surface enhanced Raman scattering prior to coating with thionine. In general very much stronger Raman signals were observed from the electrode surface than previously reported.¹⁰

(a) pH 1.3

SERR spectra of thionine, coated on gold electrodes, in 0.05 mol dm⁻³ H₂SO₄ (pH 1.3) at various applied potentials are shown in Figure 2. Also shown in this figure is the Raman spectrum from the thionine-coated dry electrode. The wavenumber shifts and the relative intensities of the bands observed in these spectra are given in Table I. Raman spectra shown in Figure 2 were recorded in sequence from A to E.

In general there is a good agreement between these Raman spectra and those reported in Ref. 10. The main difference is that in the present work much higher Raman signals (100 k counts s⁻¹ compared with 1-5 k counts s⁻¹) were obtained using the multiple oxidation reduction cycles. In the previous Raman spectroscopic study of the thionine/Au system, the Raman spectrum from the leucothionine form of the coated electrode could not be measured. Because leucothionine does not absorb at the wavelength of excitation, no resonance enhancement of the Raman spectrum occurs and there was previously very little contribution from the SER processes. In the present work Raman spectra from leucothionine could be observed, although the Raman signal was an order of magnitude lower than that from the thionine form. Since leucothionine does not absorb in the visible region of the spectrum, the Raman intensity for leucothionine is purely derived from the surface enhancement processes.

Figure 2A shows the SERRS of thionine coated on an Au electrode immediately after the coating procedure, i.e. no electrochemistry had

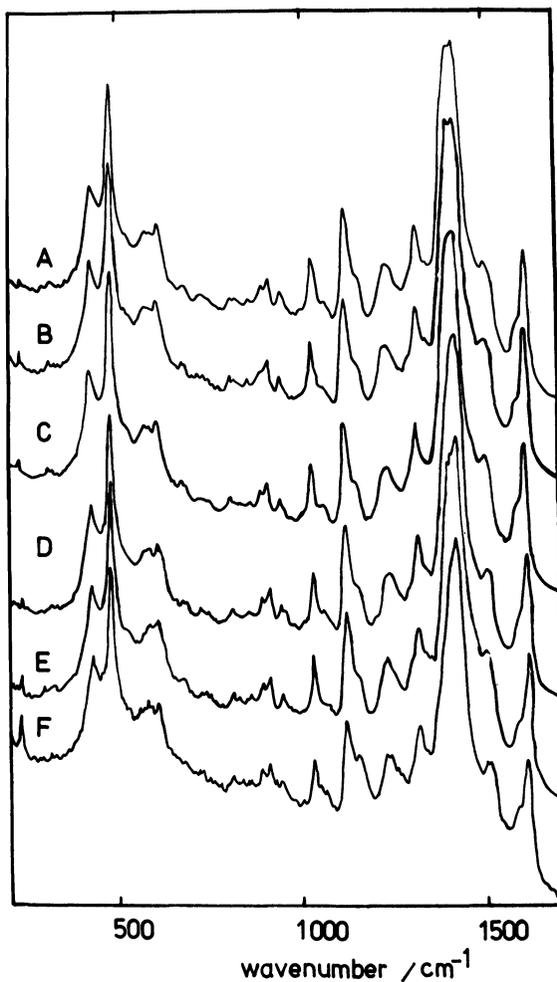


Figure 2 SERRS and SERS of thionine-coated gold electrode at pH 1.3, at various applied potentials. (A) Open circuit potential (-0.28 V) immediately after coating; (B) Spectrum recorded after measuring a cyclic voltammogram; (C) Spectrum at -0.20 V (expanded $\times 10$); (D) Spectrum at $+0.40$ V; (E) Spectrum from a dry electrode.

Table I Band wavenumbers (cm^{-1}) in SERRS and SERS of thionine-modified gold electrode at pH 1.3 at various applied potentials. 647.1 nm excitation.

Open circuit voltage (+0.28 V) immediately after coating (thionine)	Open circuit voltage (+0.28 V) after cycling between -0.2 and 0.5 V (thionine)	-0.20 V (leucothionine)	+0.40 V (thionine)	Dry electrode (thionine)
1619(s)	1619(s)	1618(sh) 1609(w) 1581(w) 1563(w)	1619(s)	1618(s)
1503(s)	1503(s)	1502(sh)	1501(s)	1500(s)
1480(sh)	1479(sh)	1490(w)	1480(sh)	1480(sh)
1426(sh)	1423(sh)	1425(sh)	1423(sh)	
1392(vs)	1393(vs)	1390(w)	1392(vs)	1391(vs)
1319(w)	1320(w)	1318(vw)	1319(w)	1314(w)
1291(w)	1292(sh)		1292(w)	1293(sh)
1223(w)	1235(w)	1240(m)	1233(vw)	1235(w)
1150(sh)	1151(m)	1152(m)	1152(m)	1150(m)
1130(m)	1132(sh)	1119	1135(sh)	1135(sh)
1038(m)	1038(m)	1036(vw)	1038(m)	1036(m)
1003(vw)	1003(w)		1003(vw)	1005(vw)
953(w)	950		955(vw)	960(w)
911(m)	911(m)	909(vw)	911(w)	909(m)
885(m)	883(sh)		888(sh)	
825(vw)	828(sh)		828(sh)	829(w-m)
807(m)	810(w)	813(w)	811(m)	803(w-m) 760(vw)
750(sh)	752(sh)		750(vw)	748(vw)
730(vw)	734(w)		735(vw)	733(vw)
681(w)	680(w)		680(w)	680(w)
616(m)	616(s)	616(m)	614(s)	614(s)
481(vvs)	481(vvs)	486(vs)	481(vvs)	481(vvs)
428(sh)	441(sh)	442(sh)	440(sh)	439(sh)
	419(sh)	420(sh)	419(sh)	418(sh)
402(m)	399(sh)	399(sh)	400(sh)	398(sh)
314(vw)	316(vw)	313(vw)	315(vw)	315(vw)
295(vw)	296(vw)		296(vw)	297(vw)

Key: s = strong, m = medium, v = very, sh = shoulder, w = weak.

been carried out with this electrode. This spectrum is in good agreement with that previously reported for thionine coated on a gold electrode.¹⁰ However, there are extra bands at *ca.* 825, 750, 730 and 295 cm^{-1} , relatively weak in intensity.

Spectrum B in Figure 2 was recorded after measuring the cyclic

voltammogram in the potential range 0.50 to -0.20 V and removing the applied voltage. Thus both spectra, 2A and B, were measured at the open circuit potential ($+0.28$ V) for the thionine coated gold electrode in contact with $0.05 \text{ mol dm}^{-3} \text{ H}_2\text{SO}_4$. There are a number of irreversible changes seen in spectrum 2B. In each case the Raman spectrum is dominated by the intense band at 480 cm^{-1} (assigned to a CNC skeletal deformation mode¹⁰). In spectrum 2A a band at 400 cm^{-1} (assigned to CSC skeletal deformation¹⁰) and a weak shoulder at 428 cm^{-1} are observed). However, in spectrum 2B there are three bands that can be clearly seen on the lower wavenumber side of the main 480 cm^{-1} band. These bands, at 440 , 419 and 399 cm^{-1} , appear as weak shoulders. The shape of the complex band structure at *ca.* 1400 cm^{-1} shows a marked difference on cycling between 0.50 and -0.20 V. For example, in spectrum 2A a shoulder at 1425 cm^{-1} to the main band at 1390 cm^{-1} appears almost as a doublet. After cycling between 0.50 and -0.20 V (spectrum 2B) there is a marked decrease in the relative intensity of this 1425 cm^{-1} shoulder. This band has been attributed to a C—N stretching vibration mode of the amino nitrogen atom bridging the two thionine molecules in the dimer formed on coating.¹⁰ The wavenumber of 1425 cm^{-1} is somewhat higher than that expected for a ν (asym) (CN) vibration mode and probably reflects some increase in the bond order and/or mode coupling. This "effect" appears to decrease on cycling the voltage of the modified electrode and hence the relative intensity of the 1425 cm^{-1} band appears to decrease.

Application of further potential and cycling does not affect the appearance of the Raman spectrum (c.f. 2B and 2D). The irreversible changes in appearance of SERRS of thionine on the gold electrode indicate that some type of reorganization of thionine layers on the surface occurs when the applied voltage is cycled after the coating procedure. This reorganization may lower the symmetry of the adsorbed thionine such that bands that were previously Raman inactive become active.

The leucothionine SER spectrum (2C) is markedly different from the SERRS of thionine. As has been stated earlier, the spectrum is an order of magnitude weaker than the SERRS of thionine under the same experimental conditions. This is attributable to the loss of the resonance enhanced contribution to the Raman intensity in the case of leucothionine. The bands that were very low in intensity in the thionine Raman spectrum are not apparent in the leucothionine

spectrum. However, there are new bands seen at *ca.* 1609, 1581 and 563 cm^{-1} . The 480 cm^{-1} band remains the most prominent feature in the Raman spectrum of leucothionine. A band at 1618 cm^{-1} appears as a weak shoulder to the main band at 1609 cm^{-1} . This whole band structure appears to be somewhat broader than the corresponding band at 1618 cm^{-1} in the thionine spectrum. The leucothionine band at 1490 cm^{-1} is to be compared with that from thionine at 1480 cm^{-1} .

Other leucothionine bands occur at 1240 and 1119 cm^{-1} . The corresponding bands in the thionine spectrum are observed at 1235 and 1130 cm^{-1} . Furthermore, a band at 1290 cm^{-1} in the Raman spectrum of thionine is absent from the leucothionine spectrum.

Spectrum D in Figure 2 was obtained on switching the potential of a thionine-coated electrode to +0.40 from -0.20 V. The overall intensity of this spectrum was similar to that observed in spectra 2A and B. All the bands present in spectrum 2B are also present in spectrum 2D, and with very similar relative intensities.

(b) pH 7

Raman spectra from a thionine-coated gold electrode, in contact with the electrolyte solution at pH 7 at applied potentials of +0.30 V, -0.20 V, and at the open circuit potential (+0.28 V) are shown in Figure 3. The sequence of recording these spectra was from A to C. The positions and the relative intensities of the bands observed in these spectra are given in Table II.

The Raman spectra from coated thionine at pH 7 are very similar to those observed at pH 1.3. Spectrum A in Figure 3 was measured at the open circuit potential. Here, bands in the 1500–1400 cm^{-1} region are at lower wavenumbers than those observed in the corresponding spectrum at pH 1.3. For example, a strong band seen at 1503 cm^{-1} in the Raman spectrum at pH 1.3 is shifted to 1499 cm^{-1} at pH 7. Similarly a band at 1425 cm^{-1} in the Raman spectrum at pH 1.3 shifts to 1420 cm^{-1} at pH 7.

At an applied potential of -0.2 V, the overall intensity of the Raman spectrum at pH 7 (spectrum B in Figure 3) goes down by an order of magnitude, just as it does in the Raman spectrum at pH 1.3. This spectrum is attributed to leucothionine. Again the band at 480 cm^{-1} is the most prominent feature in this spectrum. As at pH 1.3, additional bands at 1609, 1524 and 1350 cm^{-1} are found in the Raman spectrum of leucothionine at pH 7. These bands are not observed in

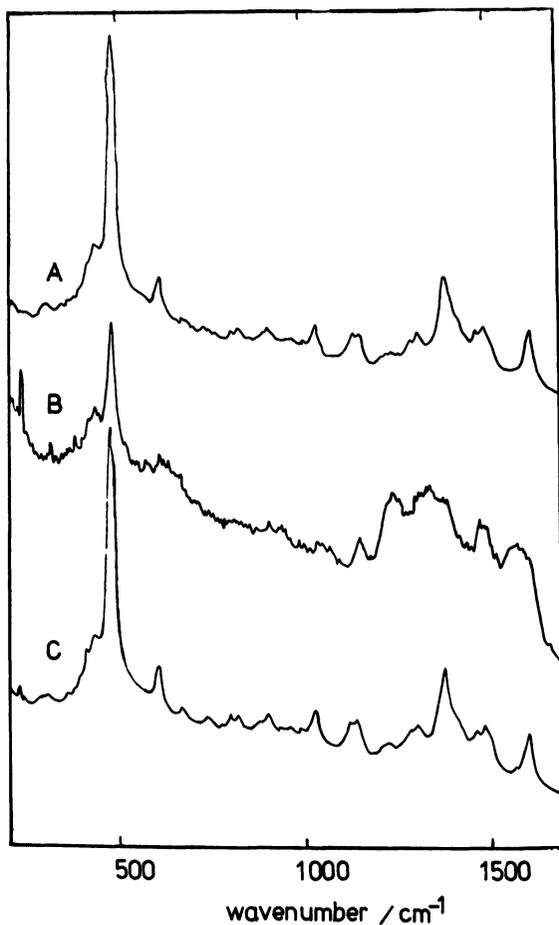


Figure 3 SERRS and SERS of thionine-coated gold electrode at pH = 7 at various applied potentials. (A) Open circuit potential (+0.28 V); (B) -0.20 V (spectrum expanded $\times 10$); (C) +0.30 V.

the spectrum of thionine at this pH. The band at 1480 cm^{-1} in the thionine spectrum at this pH is shifted to *ca.* 1490 cm^{-1} in the leucothionine spectrum. In general the bands in the $1300\text{--}1600\text{ cm}^{-1}$ region are broader and somewhat stronger in relation to the 480 cm^{-1} band in the spectrum of leucothionine at pH 7 than are those from thionine at pH 1.3. In addition, a very weak band at *ca.* 455 cm^{-1} can

Table II Band wavenumber (cm^{-1}) in SERS of thionine-modified gold electrode at pH 7 at two different potentials. Excitation wavelength 647.1 nm.

+0.30 V	-0.20 V	Open circuit (+0.28 V)
1615(s)	1615(sh)	1619(s)
	1595(sh)	
1584(vw)	1589(w)	
	1560(sh)	
	1524(sh)	
1498(s)	1500(sh)	1499(s)
1475(w-m)	1479(m)	1478(sh)
1424(sh)		1420(sh)
1388(vs)	1393(sh)	1392(vs)
1340(vw)	1350(m)	
1313(w-m)	1314(sh)	1317(w)
1290(sh)		1293(sh)
1240(w)	1245(s)	
1225(w)		
1150(m)	1156(m)	1155(m)
1132(sh)		1133(sh)
1032(m)	1040(w)	1038(m)
1003(vw)		1003(vw)
961(vw)	955(vw)	960(vw)
906(m)	909(vw)	908(w)
824(w)		830(w)
804(w)		808(sh)
757(vw)		
672(w)	670(vw)	738(vw)
		677(w)
611(s)	614(vw)	616(m)
	577(vw)	570
480(vvs)	481(vs)	481(vvs)
438(sh)	439(s)	441(sh)
417(sh)	419(sh)	416(sh)
399(sh)	402(sh)	
360(vw)		
310(vw)	312(w)	
294(vw)		298(vw)

Key: s = strong, m = medium, v = very, sh = shoulder, w = weak.

be distinguished in the Raman spectrum at pH 7. This band was not apparent in the leucothionine spectrum at pH 1.3.

(c) pH 10

The Raman spectra of a thionine coated Au electrode at pH 10 and at various applied potentials are shown in Figure 4. The wavenumbers

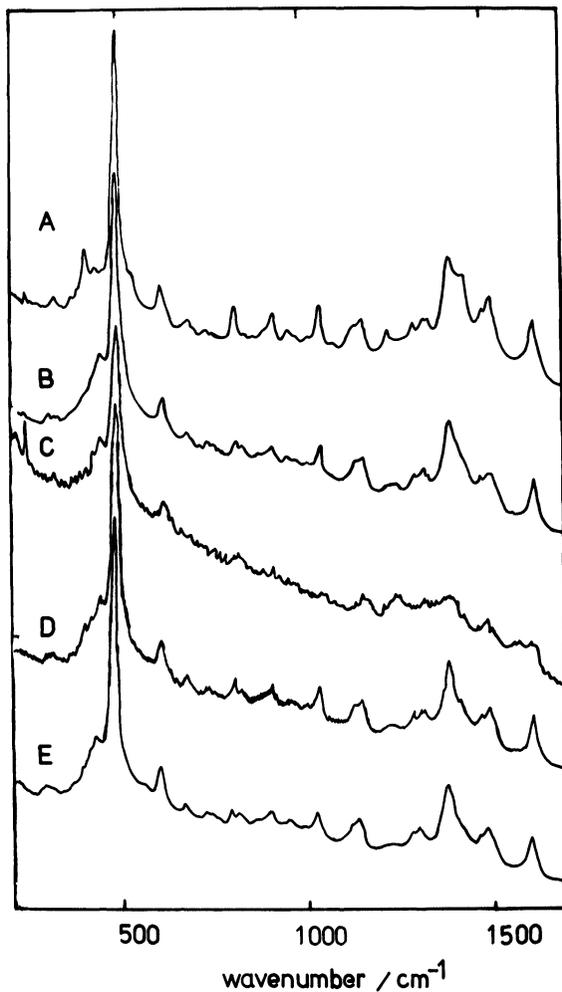


Figure 4 SERS of thionine-coated gold electrode, at pH = 10, at various applied potentials. (A) Open circuit potential (+0.27 V); (B) +0.30 V; (C) +0.10 V; (D) -0.10 V; (E) -0.30 V; (F) -0.50 V.

and the relative intensities of the bands observed in these spectra are given in Table III. As can be seen, these differ greatly from those measured at pH 1.3 and 7. Here the overall Raman signal in all the spectra is down by a factor of 10 (these spectra were recorded on the 10 k counts s^{-1} scale compared with the 100 to 200 k counts s^{-1} scale used for thionine spectra at pH 1.3 and 7). The other striking difference observed is that there was no decrease in the overall Raman signal at applied potentials of -0.30 and -0.50 V where the coated species should be in the leucothionine form (see Figure 3B).

Table III Band wavenumbers (cm^{-1}) in SERS of thionine-modified gold electrode at pH 10 at various applied potentials. Excitation wavelength, 647.1 nm.

Open circuit voltage (+27 V)	+0.30 V	+0.10 V	-0.10 V	-0.30 V	-0.50 V
1612(s)	1609(s)	1608(s)	1608(s)	1609(s)	1608(s)
1590(sh)	1589(sh)	1590(sh)	1589(sh)	1587(sh)	1587(sh)
1500(sh)	1500(sh)	1500(sh)	1500(sh)	1499(sh)	1500(sh)
1424(vvs)	1419(vs)	1419(vvs)	1419(vvs)	1424(vvs)	1420(vs)
1406(sh)	1403(vvs)			1403(sh)	
1338(vw)	1338(vw)		1337(vw)	1340(vw)	1337(vw)
1316(s)	1312(s)	1312(s)	1313(s)	1313(s)	1312(s)
1228(m)	1226(m)	1226(m)	1227(m)	1230(m)	1231(m)
1155(sh)	1151(sh)	1150(sh)	1150(sh)	1149(sh)	1147(sh)
1121(s)	1118(s)	1116(s)	1117(s)	1119(s)	1116(s)
1062(w)	1060(w)	1060(w)	1058(w)	1062(w)	1056(w)
1030(m)	1026(m)	1027(m)	1027(m)	1027(m)	1022(m)
1003(vw)		1004(vw)	1003(vw)	1003(vw)	
946(w)	940(w)	940(w)	940(w)	940(w)	937(vw)
911(w)	908(w)	908(w)	908(w)	908(w)	908(w)
890(sh)	888(sh)	888(sh)	887(w)	891(w)	885(sh)
855(vw)	854(vw)	855(vw)	851(vw)	854(vw)	855(vw)
830(vw)				828(vw)	
808(vw)	807(vw)	806(vw)	805(w)	808(vw)	806(w)
746(vw)		746(vw)	742(vw)	744(vw)	
722(vw)	721(vw)	720(vw)	722(vw)	723(vw)	725(vw)
680(w)	681(w)	680(w)	680(w)	675(w)	680(vw)
612(s)	616(s)	609(s)	609(s)	608(s)	608(m)
579(sh)	580(sh)	582(sh)	580(sh)	581(sh)	580(sh)
481(vs)	479(vs)	478(vs)	480(vs)	478(vvs)	478(vvs)
				448(sh)	
427(s)	426(s)	424(s)	425(s)	425(s)	424(s)
313(vw)	310(vw)	312(vw)	312(vw)	310(vw)	310(vw)
292(vw)				290(vw)	

Key: s = strong, m = medium, v = very, sh = shoulder, w = weak.

At pH 10 the most prominent feature in the Raman spectra of coated thionine is no longer observed at 480 cm^{-1} , as was the case at pH 1.3 and 7. Here, all the spectra are dominated by the complex band structure centred at *ca.* 1420 cm^{-1} . This band appears as an unresolved doublet at the open circuit potential ($+0.27\text{ V}$, spectrum A in Figure 4) with components at 1424 and 1406 cm^{-1} . At an applied potential of $+0.30\text{ V}$ (spectrum B in Figure 4) this band also has a doublet structure with peaks centred at 1419 and 1403 cm^{-1} . On changing the applied potential to $+0.10\text{ V}$ (spectrum C in Figure 4) only a broad band centred at 1419 cm^{-1} is apparent. Similarly when the applied potential is switched to 0.10 V (spectrum D in Figure 4) again only one band, centred at 1419 cm^{-1} , is observed. At an applied potential of -0.30 V again a shoulder appears at 1403 cm^{-1} , with the main band centred at 1424 cm^{-1} .

At pH 1.3 and 7, there are two bands, at 1390 and 1425 cm^{-1} , observed in the Raman spectra of thionine. The band at 1390 cm^{-1} also appears in the resonance Raman spectrum of solid thionine and has been assigned to the asymmetric CN stretching mode of the thionine molecule. The band at 1425 appears only in the Raman spectra of coated thionine and has been attributed to the asymmetric stretching vibration of the CNC bond bridging the two thionine molecules in the dimer formed on the electrode surface. These wavenumber shifts for the asymmetric C—N stretching vibrational mode are higher than those expected for secondary amines¹⁵ and may be explained on the basis of the higher bond order and/or some mode mixing.¹⁰

At pH 10 the 1390 cm^{-1} band shifts up to 1400 – 1420 cm^{-1} . This shift suggests an increase in the CN double bond character in the thionine molecule.

The band at 1618 cm^{-1} , assigned to a CC stretching vibration¹⁰ in the Raman spectra of coated thionine at pH 1.3 and 7, shifts down to 1612 cm^{-1} on changing the pH of the supporting electrolyte solution to 10, and a shoulder at *ca.* 1590 cm^{-1} is apparent at this pH. This 1590 cm^{-1} band is not apparent in the Raman spectra of thionine at pH 1.3 and 7, but there is a band seen at 1581 and 1589 cm^{-1} in the spectra of leucothionine at these pH values. At pH 10, the 1612 cm^{-1} band shifts to 1609 cm^{-1} on applying a potential of $+0.30\text{ V}$ but does not shift at other values of applied potential.

There is a marked increase in the relative intensity of the band at 1312 – 1316 cm^{-1} in the spectrum of coated thionine at pH 10. This band

appears as a weak feature at $1313\text{--}1320\text{ cm}^{-1}$ in the spectra at pH 1.3 and 7. There is an additional weak band which appears at 1340 cm^{-1} in the spectrum at pH 10. The band at $1226\text{--}1230\text{ cm}^{-1}$ shows a marked increase in relative intensity compared with the similar band at $1223\text{--}1240\text{ cm}^{-1}$ seen in the Raman spectra at pH 1.3 and 7.

There are changes in the relative intensities and positions of two bands observed in $1100\text{--}1150\text{ cm}^{-1}$ region of the Raman spectrum on changing the pH of the electrolyte solution to 10. At pH 1.3 and 7 the two bands are observed at *ca.* 1130 and 1150 cm^{-1} with the latter band more intense than the former. However, at pH 10 the dominant band appears at $1116\text{--}1120\text{ cm}^{-1}$ with a shoulder at $1147\text{--}1155\text{ cm}^{-1}$. Again there is a marked increase in the relative intensities of these two bands compared with those of the corresponding bands at pH 1.3 and 7. The band at 1130 cm^{-1} in the spectrum of coated thionine at pH 1.3 has been assigned to a S = O vibration resulting from the oxidation of the S atom of thionine during the electro-coating process.¹⁰ A new band at 1060 cm^{-1} is observed in the spectrum of coated thionine at pH 10. This band was not apparent in the spectra at pH 1.3 or 7. The band in the region $1023\text{--}1030\text{ cm}^{-1}$ at pH 10 is at a lower wavenumber than the corresponding band of thionine at pH 1.3 and 7, i.e. $1036\text{--}1040\text{ cm}^{-1}$. Similarly there is a shift to lower wavenumber of the band observed at $950\text{--}980\text{ cm}^{-1}$ at pH 1.3 and 7. At pH 10 this band appears at $937\text{--}946\text{ cm}^{-1}$.

Other major differences in the Raman spectra of coated thionine at pH 10 are observed in the $400\text{--}500\text{ cm}^{-1}$ region. Here, as in the case of the spectra at pH 1.3 and 7, the strongest band is at 480 cm^{-1} . However, only a single strong and relatively broad band is observed at $424\text{--}427\text{ cm}^{-1}$. In the Raman spectra of thionine at pH 1.3 and 7, at least three bands, at *ca.* 440 , 420 and 400 cm^{-1} , were apparent. There is a marked increase in relative intensity of the band at $424\text{--}427\text{ cm}^{-1}$ at pH 10.

The large changes in band wavenumbers and intensities observed in the Raman spectra on raising the pH of the electrolyte solution to pH 10 suggest major changes in both the molecular structure and the absorption spectrum of coated thionine. The decrease in the overall Raman signal suggests some loss of resonance enhancement which could arise from a shift in the absorption spectrum. As with the leucothionine spectra, the spectra obtained at pH 10 appear to be attributable mainly to SERS effects.

Thionine in solution exists in various protonated forms^{16,17} depending on the pH. For example, at pH < 2 thionine exists in its diprotonated form, which absorbs strongly at 673 nm. In the pH range 2 to 10, the monoprotonated form is dominant with a strong absorption at 597 nm. In strongly alkaline solution (5% NaOH) a basic form¹⁶ exists, with an absorption band at 515 nm. The transition between the monoprotonated form and thionine hydroxide is reported to occur at pH 11.¹⁶

The Raman spectra of the thionine modified gold electrode at pH 10 may be attributed to thionine hydroxide. The transition from protonated thionine to thionine hydroxide on the electrode surface appears to take place at lower pH than that reported for the solution species, but no absorption data are available in the literature for thionine modified electrodes at different pH values.

Acknowledgements

We are grateful to the SERC for support of this work.

References

1. W. J. Albery and A. R. Hillman, Annual Reports C75 (The Royal Society of Chemistry, London, 1981) p. 377.
2. R. W. Murray, *Phil. Trans. Roy. Soc. London. Ser. A* **302**, 253 (1981).
3. W. J. Albery, *Acc. Chem. Res.* **15**, 142 (1982).
4. W. J. Albery, A. W. Faulds and A. R. Hillman, *J. Electrochem. Soc.* **127**, 654 (1980).
5. W. J. Albery, W. R. Bowen, F. Fisher, A. W. Faulds, K. J. Hall, A. R. Hillman, R. G. Edgell and A. R. Orchard, *J. Electroanal. Chem.* **107**, 37 (1980).
6. W. J. Albery, A. W. Faulds, K. J. Hall, A. R. Hillman, R. G. Edgell and A. F. Orchard, *Nature* **282**, 793 (1979).
7. J. M. Boulderey and M. D. Archer, *Electrochim. Acta* **28**, 1575 (1983).
8. W. J. Albery, M. G. Boutelle, P. J. Colby and A. R. Hillman, *J. Electroanal. Chem.* **133**, 135 (1982).
9. W. J. Albery, A. R. Hillman, R. G. Edgell and M. Nutton, *J. Chem. Soc. Faraday Trans. 1* **80**, 111 (1984).
10. K. Hutchinson, R. E. Hester, W. J. Albery and A. R. Hillman, *J. Chem. Soc. Faraday Trans. 1* **80**, 2053 (1984).
11. P. Gao, M. L. Patterson, M. A. Tadyyoni and M. J. Weaver, *Langmuir* **1**, 173 (1985).
12. P. Gao and M. J. Weaver, *J. Phys. Chem.* **89**, 5040 (1985).
13. M. L. Patterson and M. J. Weaver, *J. Phys. Chem.* **89**, 5046 (1985).
14. H. R. Virdee and R. E. Hester, *Croat. Chim. Acta*, **61**, 355 (1988).
15. F. R. Dollish, W. G. Fately and F. F. Bentley, *Characteristic Raman Frequencies of Organic Compounds*, Wiley-Interscience, New York, 1974.
16. L. F. Epstein, F. Karush and E. Robinowitch, *J. Opt. Soc. Am.* **31**, 77 (1941).
17. E. Rabinowitch and L. F. Epstein, *J. Am. Chem. Soc.* **63**, 69 (1941).
18. E. Rabinowitch, *J. Chem. Phys.* **8**, 551 (1940).