ORIGINAL PAPER

The adsorption and faradaic processes of formylferrocene thiosemicarbazone monitored by in situ SERS and UV-VIS spectroscopies

Gustavo F. S. Andrade · Marcia L. A. Temperini

Abstract The adsorption and faradaic processes of formylferrocene thiosemicarbazone (TFF) on gold electrode in 0.1 mol L^{-1} $NaClO_4$ acetonitrile solution were monitored by surface-enhanced Raman scattering (SERS) and ultraviolet-visible (UV-VIS) techniques. SERS data indicate that TFF adsorbs through the iminic N(2′) and S atoms on the gold electrode. The reduction product formed on the gold surface was aminomethylferrocene, whose experimental spectrum was supported by density functional theory calculations. In solution, thiourea was detected by the UV-VIS technique. Although there was an oxidation wave in the TFF cyclic voltammogram, no spectral changes were observed after the oxidation process.

Keywords Formylferrocene thiosemicarbazone ·
Faradaic reactions · Surface-enhanced Raman scattering ·
UV-visible spectroscopy · Density functional theory

Introduction

Thiosemicarbazone derivatives are a family of compounds with great interest because of their biological properties as antibacterial, antimalarial, antineoplastic, and antiviral drugs [1–6]. Some of these compounds are also surface modifiers of gold electrodes, allowing the study of electrochemical properties of biological molecules, such as horse heart cit c [7]. The knowledge of the modifiers' adsorption configura-tion and faradaic behavior in metallic surfaces is thus important for understanding the structure/properties relation.

The reduction behavior of thiosemicarbazones has been widely studied using electrochemical techniques, and Soucaze-Guillous and Lund [8] proposed a general pathway to the reduction of this family of compounds. In a recent work, it has been shown using surface-enhanced Raman scattering (SERS) and ultraviolet-visible (UV-VIS) techniques that 2-formylpyridine thiosemicarbazone follows this general reduction pathway, resulting in 2-picolyl-amine and thiourea as reduction products [9].

Formylferrocene thiosemicarbazone (TFF, structure in Scheme 1) has the thiosemicarbazone moiety and a ferrocenyl group. Ferrocene derivatives have been widely studied by electrochemical techniques and are employed as amperometric glucose biosensors [10, 11]. TFF and some of its derivatives were studied by cyclic voltammetry [12] and show a reversible process with $E_{1/2}=0.56$ V vs saturated calomel electrode assigned to Fe^{II}/Fe^{III}.

The vibrational spectrum of ferrocene has been intensively studied [13–20], and the bands assignments are well established. In a recent work, the resonance Raman study of TFF was reported, and a band assignment based on the density functional theory (DFT) approach was proposed [21].

The aim of this work is to characterize the adsorption of TFF and its faradaic reduction and the oxidation processes on gold surfaces using in situ SERS and UV-VIS absorption techniques.

Experimental section

Chemicals

Acetonitrile (Aldrich) was distilled before use. Deionized water was used to prepare all the aqueous solutions. The salts

---

This work is dedicated to the memory of Prof. Francisco C. Nart.

G. F. S. Andrade · M. L. A. Temperini (✉)
Instituto de Química, Universidade de São Paulo,
Caixa Postal 26077,
05513-970 São Paulo, SP, Brazil
e-mail: mlatempe@iq.usp.br

Scheme 1 Structure of TFF
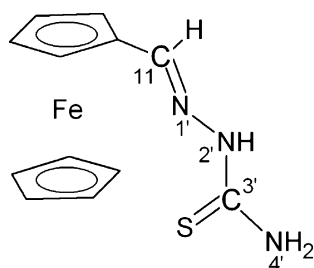
KCl (Fluka) and NaClO$_4$ (Merck) were used as received. Thiosemicarbazide was recrystallized from 1:1 vol water/ethanol, and formylferrocene was used as received.

Synthesis of TFF

TFF was synthesized using a method described in the study of Wiles and Suprunchuk [22]: 25 ml of an ethanolic solution of formylferrocene (265 mg, 1.25 mmol) was added dropwise to 30 ml of an aqueous solution of thiosemicarbazide (114 mg, 1.25 mmol). Acetic acid (0.5 ml) was added to this solution that was heated at 70 °C for 20 min. The orange solid formed was filtered off and washed with 15 ml of a cold 1:1 vol water/ethanol solution and dried under vacuum. The results of the elemental analysis (expected/calculated) were the following: C= 50.2%/50.3%, H=4.56%/4.68%, and N=14.6%/14.8%.

Treatment of the metallic surface and self-assembly procedure for SERS experiments

Before each experiment, the working electrode was mechanically polished with a 600-mesh sandpaper, rinsed with water, and then polished with a 1,000-mesh sandpaper. For cleaning the electrode, it was sonicated for 10 min in ethanol, rinsed with deionized water, and sonicated for 10 min in deionized water.

The SERS activation was performed by 25 successive oxidation–reduction cycles applied to the electrode in a 0.1 mol L^{-1} KCl aqueous solution in the absence of TFF. After the roughening procedure, the activated electrode was thoroughly rinsed with pure acetonitrile. The self-assembly was done by immersion of the electrode in a saturated solution of TFF (about 70 mmol L^{-1}) in acetonitrile for 20 h. The electrode was then rinsed with pure acetonitrile. For the in situ procedures, the electrode was subsequently placed in the electrochemical cell filled with a solution of 0.1 mol L^{-1} NaClO$_4$ in acetonitrile, saturated with TFF to acquire the SERS spectra. In the ex situ procedure, the electrode was dried, and the SERS spectra were obtained from the dry surface.

The SERS and Raman (of the solid compound) spectra were obtained in a Renishaw Raman Imaging Microscope System 3000 fitted with a Peltier-cooled charge-coupled device detector and with an Olympus metallurgical microscope with a 50× objective lens to focus the laser beam on the sample. As exciting radiation, the 632.8-nm line from an air-cooled He–Ne laser (Spectra Physics model 127-35) was used. The laser power at the sample was approx. 100 μW to avoid sample decomposition.

Cyclic voltammetry

The cyclic voltammograms were obtained using an EG&G Princeton Applied Research model PAR-263A Potentiostat/Galvanostat. The TFF concentration used was 1 mmol L^{-1} in a 0.1-mol L^{-1} NaClO$_4$ acetonitrile solution. The working electrode was a 0.13-cm^2 polycrystalline gold rod (99.99%) inserted in a Teflon sleeve; the auxiliary and reference electrodes were a platinum wire and an Ag wire, respectively.

In situ UV-VIS spectra

The UV-VIS spectra were obtained on a HP8453 spectrophotometer and with an EG&G Princeton Applied Research model PAR-362 Potentiostat/Galvanostat. The working solution was 5.0×10^{-4} mol L^{-1} TFF in 0.1 mol L^{-1} NaClO$_4$ acetonitrile. The spectroelectrochemical cell consists of a standard quartz cuvette with an optical path of 10 mm, where the working (gold wire), reference (silver wire), and counter electrodes (platinum wire) are inserted in a Teflon sleeve close to the walls, allowing a solution-free volume for detection, as can be seen in Scheme 2. As the reduced dimensions of the cuvette did not allow stirring, the potentials were applied during 5 min before each spectra.

Computational details

The equilibrium structure and vibrational wavenumber of aminomethylferrocene were calculated by the DFT ap-
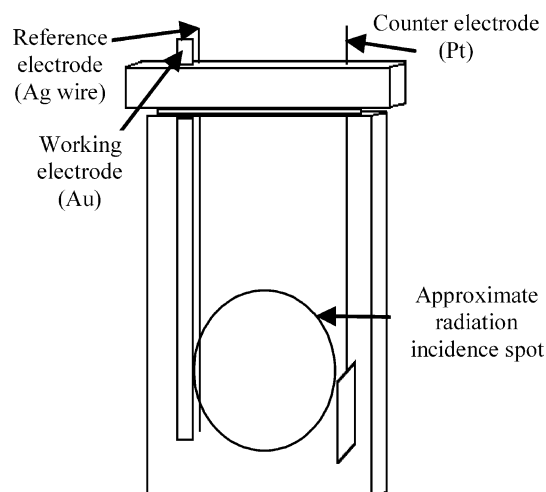
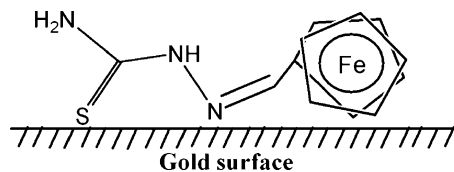**Scheme 2** Spectroelectrochemical cell for the in situ UV-VIS experiments
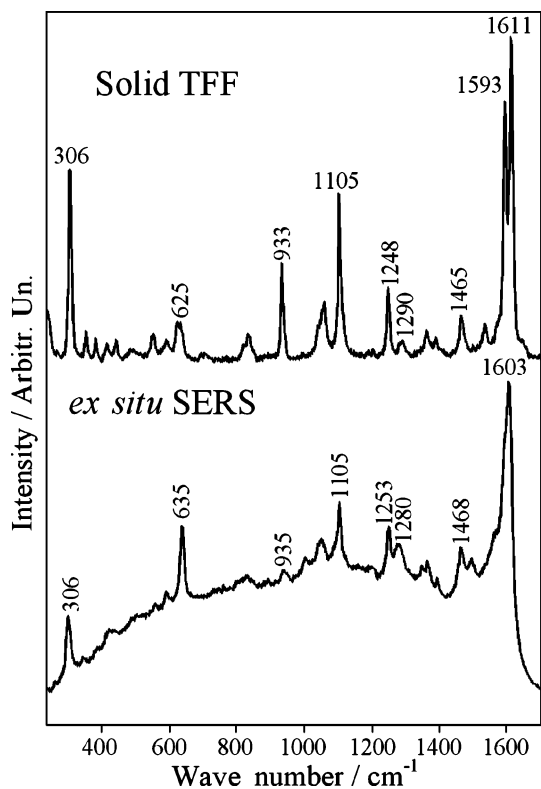
Fig. 1 Raman spectrum of TFF in the solid state and ex situ SERS spectrum of a dry gold electrode with TFF adsorbed from an acetonitrile solution

Scheme 3 Proposed adsorption configuration of TFF on gold electrodes

agrees well with the configuration of TFF acting as a ligand in gold complexes [26]. Using the golden rule of SERS, which states that the bands assigned to moieties bonded directly to the surface are the most enhanced [27], the increase in the relative intensities of the bands assigned to the thiosemicarbazone group in the SERS spectrum is

proach using the B3LYP functional [23, 24] and the double-ζ quality with polarization function 6-31G(d) basis set from the Gaussian 98 program package [25] following the same procedure used in TFF calculations [21].
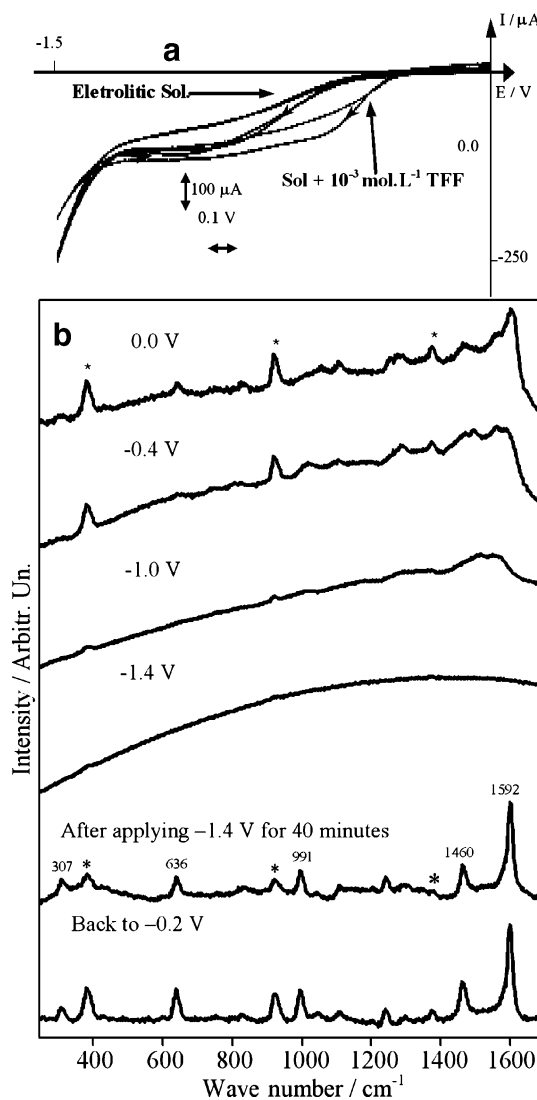
Results and discussion

The ex situ SERS spectrum of TFF on the gold electrode and the Raman spectrum of the compound in the solid state are shown in Fig. 1. Comparing the two spectra, it can be seen that the strongest ferrocenyl bands (306 and 1,105 cm^{-1}) do not shift upon adsorption, while the thiosemicarbazone moiety bands present considerable wavenumber shifts. Indeed, the normal Raman spectrum of the compound presents intense bands in 1,593 and 1,611 cm^{-1}, assigned respectively as $\nu(C = N^{1'})$ and $\delta(N^{4'}H_2)+\nu(C=N^{1'})$ [21], and in the SERS spectrum, one can observe only one band at 1,599 cm^{-1}. In the normal Raman spectrum, the bands at 625 and 1,290 cm^{-1}, assigned to $\delta(N^{2'}CN^{4'})$ and to $\nu(CN^{4'})+\delta(N^{2'}H)+\delta(C^{11}H)$, respectively, are observed at 635 and 1,278 cm^{-1} with higher relative intensity in the SERS spectrum. The wavenumber shifts of these bands indicate that TFF interacts with the metallic electrode through the thiosemicarbazone moiety groups, primarily by the $N^{1'}$ and S atoms (see Scheme 3). This adsorption configuration

Fig. 2 a Cyclic voltammogram of a gold electrode with TFF (1.0 mmol L^{-1}) in 0.1 mol L^{-1} NaClO$_4$ acetonitrile solution for negative potentials; sweep rate=100 mV s^{-1}. b In situ SERS spectra of TFF adsorbed in a gold electrode in NaClO$_4$ 0.1 mol L^{-1} in acetonitrile solution for several applied potentials. Asterisk, acetonitrile bands

Table 1 Experimental wavenumber in the TFF SERS spectrum at $E=0.0$ and -1.4 V, characteristic frequencies of the amino group [28], the calculated frequencies of aminomethylferrocene using the DFT approach, and a frequency assignment

SERS (TFF, 0.0V)	SERS (reduction product)	CH$_2$–NH$_2$ group wavenumber [28]	DFT wavenumber[a]	Assignment[b]
306	307		329	ν(FeCp$_2$)
639	636		667	β(CCC)+β(NH)
	827	750–850	853	ω(NH$_2$)
	991	1,035–1,045	1,012	ν(CN)
1,104	1,103		1,060	ν_s(CC) Cp
	1,199		1,105	ν(CN)+ν_s(CC) Cp
1,251	1,236		1,232	ν_s(CC) Cp (1)+ν(CC^{11})
1,276	1,292		1,314	τ(NH$_2$)
1,465	1,460		1,429	ν_a(CC) Cp(1)
1,599	1,592	1,590–1,650	1,695	δ(NH$_2$)

[a] Calculated for aminomethylferrocene; the calculated wavenumbers were multiplied by an adjust factor of 0.9804 [33]. All wavenumbers are reported in cm^{-1}.

[b] ν Stretching; δ in-plane bending; β out-of-plane bending; ρ rocking; ω wagging

experimental evidence that reinforces the proposed adsorption configuration for TFF in gold electrodes.

For negative potentials, the voltammogram of the gold electrode in 1 mmol L^{-1} TFF in 0.1 mol L^{-1} NaClO$_4$ acetonitrile solution (Fig. 2a) shows a cathodic current at potentials more negative than -0.6 V, indicating that a TFF reduction process occurs. No anodic current is observed in the reversal sweep, indicating that the reduction is an irreversible process. In Fig. 2b, the in situ SERS spectra of TFF adsorbed on a gold electrode in 0.1 mol L^{-1} NaClO$_4$ acetonitrile solution at different applied potentials are presented. In these spectra, the relative intensity of the band at 1,599 cm^{-1} decreases as the potential becomes more negative. For potentials more negative than -1.0 V, no TFF bands can be observed, and the S/N ratio markedly decreases.

Although the CV curve indicates a TFF reduction process, the SERS spectra obtained for potentials more negative than -1.0 V (Fig. 2) show only weak solvent bands. Nevertheless, after applying -1.4 V for 40 min, yellow regions can be seen on the gold electrode, and a different SERS spectrum is obtained on the entire electrode surface. This spectrum is different than the SERS spectrum of TFF at 0.0 V and presents a higher S/N ratio. This spectrum can be attributed to the reduction product. The spectrum remains the same when the potential is turned back to -0.2 V (Fig. 2b), confirming the irreversible character of the reduction process observed by CV. The new bands at 827, 991, 1,199, and 1,592 cm^{-1} can be assigned to vibrational modes of the amine group [28], while the bands at 307, 636, 1,103, and 1,460 cm^{-1}, which have corresponding bands in the SERS spectrum before reduction, are characteristic of the ferrocenyl moiety [13–20]. This spectral pattern leads one to suggest that the reduction product is aminomethylferrocene.

To verify if the spectrum obtained is of aminomethylferrocene, calculations of the wavenumbers of the vibrational bands of this compound were done using the DFT approach. In Table 1, the calculated wavenumbers are
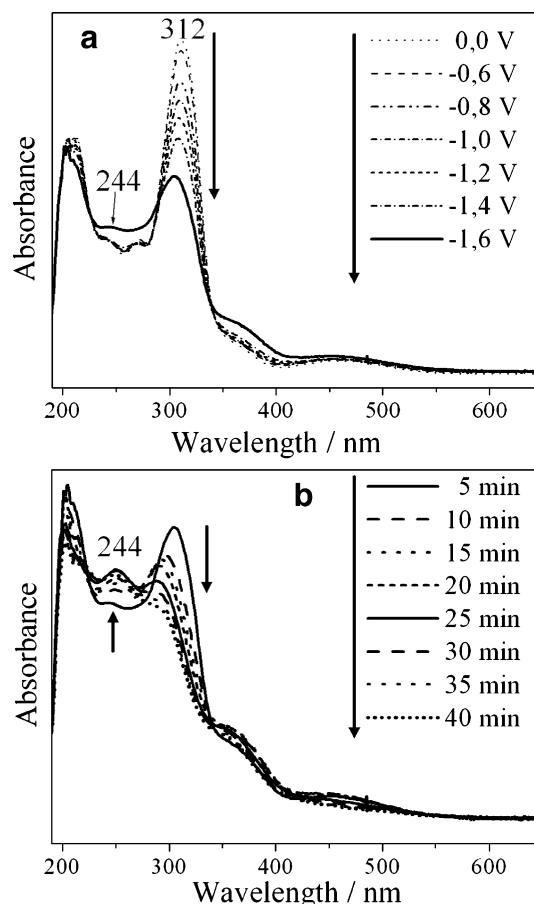
Fig. 3 a In situ UV-VIS spectra of TFF (5.0×10^{-4} mol L^{-1}) in 0.1 mol L^{-1} NaClO$_4$ acetonitrile solution; **b** In situ UV-VIS spectra of TFF with $E=-1.4$ V as a function of time
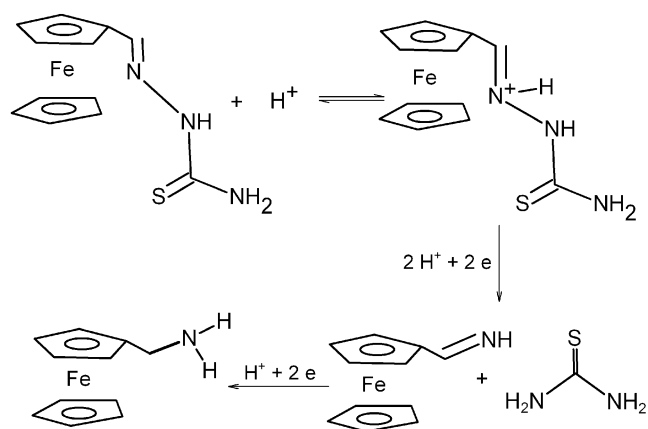
Scheme 4 TFF reduction pathway [8] as indicated by SERS and UV-VIS techniques: Aminomethylferrocene and thiourea are the reduction products

presented together with the experimental wavenumbers of TFF and the reduction product. As can be seen, there is a high coincidence between the calculated spectrum of aminomethylferrocene and the SERS spectrum of the reduction product at −1.4 V, which is another evidence that aminomethyferrocene is formed.

To further evaluate the faradaic reduction process of TFF, in situ UV-VIS was used. Figure 3 presents the in situ UV-VIS spectra of the compound as a function of the applied potential in an acetonitrile electrolytic solution, having 1.0 mmol L^{-1} of TFF in 0.1 mol L^{-1} NaClO$_4$. At 0.0 V, the spectrum presents a strong band at 312 nm, assigned to the $n-\pi^*$ transition of the thiosemicarbazone moiety, involving a molecular orbital mainly centered in the C = N$^{1'}$ and C=S groups [29, 30]. At potentials more negative than −0.8 V (Fig. 3a), the band at 312 nm looses intensity, the bands at 220 and 255 nm coalesce, and a weak band at 244 nm appears. These spectral changes indicate that a reduction process is occurring.

In Fig. 3b, the evolution of the UV-VIS spectra at −1.6 V in function of time is shown. In these spectra, the thiosemicarbazone moiety band at 312 nm decreases, and a new band at 244 nm increases for longer times. The band at 244 nm can be assigned to thiourea, because this compound has a strong band at 240 nm [9].

Based on SERS and UV-VIS data, the reduction products of TFF are aminomethylferrocene and thiourea, which is in agreement with the reaction pathway proposed by Soucaze-Guillous and Lund [8]. Based on this mechanism, the reduction of an aromatic thiosemicarbazone should occur through the transference of four electrons:

Fig. 4 a Cyclic voltammograms of a gold electrode in TFF (1.0 mmol ▶ L^{-1}) in 0.1 mol L^{-1} NaClO$_4$ acetonitrile solution with sweep rates: $a=$ 10; $b=20$; $c=30$; $d=50$; $e=100$; $f=200$ mV s^{-1}. **b** In situ SERS spectra of TFF on a gold electrode in NaClO$_4$ 0.1 mol L^{-1} acetonitrile solution for positive potentials. *Asterisk*, solvent bands. **c** SERS spectrum after applying $E=0.0$ V for 40 min. **d** In situ UV-VIS spectra of TFF (5.0×10^{-4} mol L^{-1}) in the positive potentials range
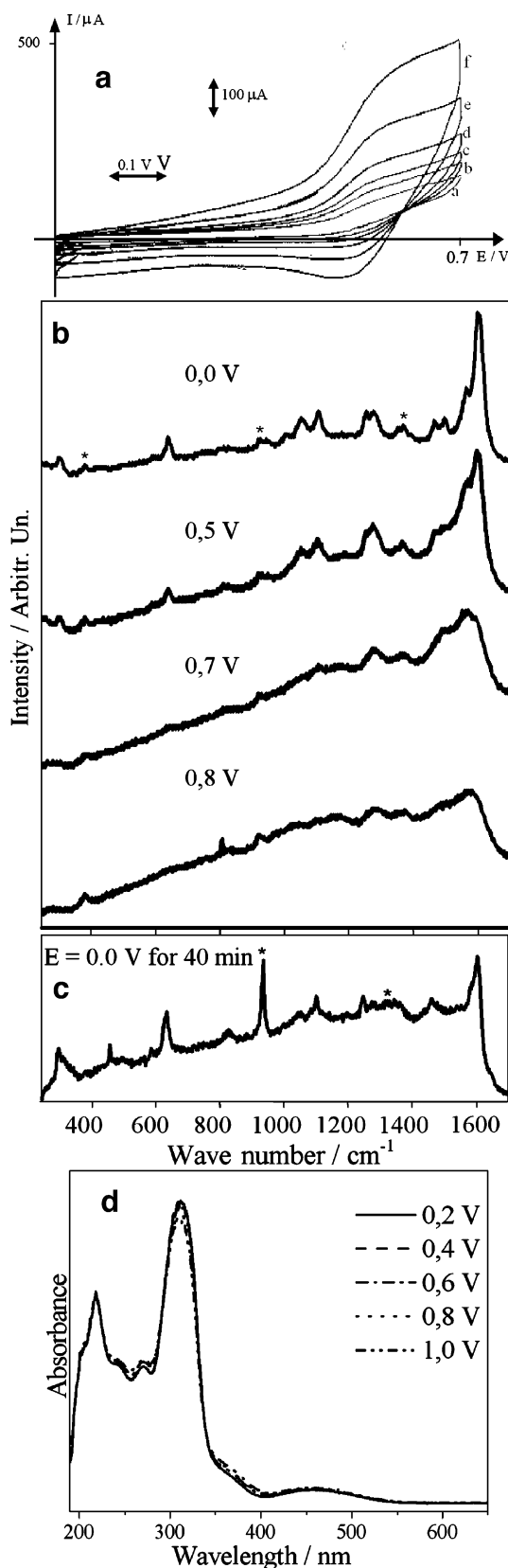
the cleavage of the N–N single bond by reduction with two electrons and then reduction by two electrons at the imine formed, as shown in Scheme 4.

The SERS and UV-VIS techniques are also used for monitoring the oxidation of the ferrocenyl moiety of TFF. This process was observed at 0.53 V in the cyclic voltammogram of TFF in a 0.1 mol L^{-1} Bu_4NClO_4 acetonitrile solution [12]. Figure 4a presents the CV curves of TFF in a 0.1 mol L^{-1} $NaClO_4$ acetonitrile solution in the positive potential range for the indicated sweep rates. The CV curves and peak values correspond to a reversible process, and they agree well with those in the study of Graúdo et al. [12]. The SERS spectra of TFF at positive applied potentials are presented in Fig. 4b. In these spectra, the relative intensity of the bands of TFF and the S/N ratio are practically constant from 0.0 to 0.4 V applied potentials. At more positive potentials, the signal decreases and the spectrum at 0.8 V shows only weak solvent bands and a broad feature from 1,200 to 1,600 cm^{-1}, because of amorphous carbon.

The decrease in the S/N ratio of the spectra after oxidation of TFF can be due to desorption of the oxidized compound, because it has a positive charge and can suffer electrostatic repulsion by the electrode surface. Another possible explanation to this result is the decomposition of the oxidized compound, as the ferricenyl group is a more reactive species than the ferrocenyl moiety [31]. No bands are observed in the spectrum at 0.0 V after applying 0.8 V to the electrode, but if 0.0 V is applied for 40 min, the SERS spectrum obtained (Fig. 4c) is very similar to the initial spectrum at 0.0 V but with lower S/N ratio.

The in situ UV-VIS spectra of TFF at positive potentials did not present any significant change as a function of applied potential, even for potentials as positives as 1.0 V, as can be seen in Fig. 4d. The behavior of TFF is different from those of other ferrocene derivatives, which suffer a shift of the d–d electronic transition in the visible region to lower energy [32]. A possible explanation to this behavior is that TFF shows a strong electronic delocalization, as has been verified by resonance Raman spectroscopy [21], diminishing the shift of d–d electronic transition.

Conclusions

In this work, from the ex situ SERS results, it was proposed that the TFF adsorption on the gold electrode occurs through the S and iminic $N^{1'}$ atom.

The electrochemical reduction process of TFF was investigated. The necessity of the association of two different techniques for a complete understanding of the reduction process was demonstrated. It was verified that the faradaic reduction products of TFF are aminomethylferrocene, observed in the in situ SERS spectra, and thiourea, detected in the in situ UV-VIS spectra. Several new bands observed in the SERS spectrum of the reduction product have their counterpart in the aminomethylferrocene vibrational spectrum obtained by DFT calculations.

The SERS spectra at positive potentials, close to the oxidation of the ferrocenyl moiety (0.53 V), did not show any band that could be assigned to the oxidized TFF.

References

1. Wiles DM, Suprunchuk T (1971) J Med Chem 14:252
2. French FA, Blanz EJ Jr, Shaddix SC, Brockman RW (1974) J Med Chem 17:172
3. Beraldo H (2004) Quim Nova 27:461
4. Jeragh BJA, El-Dissouky A (2005) J Coord Chem 58:1029
5. Mendes IC, Moreira JP, Speziali NL, Mangrich AS, Takahashi JA, Beraldo H (2006) J Braz Chem Soc 17:1571
6. Dodoff NI, Kovala-Demertzi D, Kubiak M, Kuduk-Jaworska J, Kochel A, Gorneva GA (2006) Zeitsch Naturforsch B-J Chem Sci 61:1110
7. Hill HAO, Page DJ, Walton NJ (1986) J Electroanal Chem 208:395
8. Soucaze-Guillous B, Lund H (1997) J Electroanal Chem 423:109
9. Pessôa MMB, Andrade GFS, dos Santos MR, Temperini MLA (2003) J Electroanal Chem 545:117
10. Cass AGE, Davis G, Francis GD, Hill HAO, Aston WJ, Higgins IJ, Plotkin EV, Scott LDL, Turner APF (1984) Anal Chem 56:667
11. Badia A, Carlini R, Fernandez A, Battaglini F, Mikkelsen SR, English AM (1993) J Am Chem Soc 115:7053
12. Graúdo JEJC, Filgueiras CAL, Marques-Netto A, Batista AA (2000) J Braz Chem Soc 11:237
13. Lippincott ER, Nelson RD (1958) Spectrochim Acta 10:307
14. Sado A, West R, Fritz HP, Schafer L (1966) Spectrochim Acta 22:509
15. Long TV Jr, Huege FR (1968) Chem Commun 1968:1239
16. Bodenheimer JS, Loewenthal E, Low W (1969) Chem Phys Lett 3:715
17. Bodenheimer JS, Low W (1973) Spectrochim Acta A 29:1733
18. Bailey RT (1971) Spectrochim Acta A 27:199
19. Aleksanyan VT (1982) In: Durig JR (ed) Vibrational spectra and structure. Elsevier, Amsterdam, p 107
20. Diana E, Rossetti R, Stanghellini PL, Kettle SFA (1997) Inorg Chem 36:382
21. Andrade GFS, Siqueira LJA, Ribeiro MCC, Sala O, Temperini MLA (2006) J Raman Spectrosc 37:498
22. Wiles DM, Suprunchuk T (1968) Can J Chem 46:1865
23. Becke AD (1993) J Chem Phys 98:5648
24. Lee C, Yang W, Parr RG (1988) Phys Rev B 37:785
25. Frisch MJ, Trucks GW, Schlegel HB, Scuseria GE, Robb MA, Cheeseman JR, Zakrzewski VG, Montgomery JA, Stratmann RE, Burant JC, Dapprich S, Millan JM, Daniels AD, Kudin KN, Strain MC, Farkas O, Tomasi J, Barone V, Cossi M, Cammi R, Mennucci B, Pomelli C, Adamo C, Clifford S, Ochterski J,

Petersson GA, Ayala PY, Cui Q, Morokuma K, Malick DK, Rabuck AD, Raghavachari K, Foresman JB, Cioslowski J, Ortiz JV, Stefanov BB, Liu G, Liashenko A, Piskorz A, Komaromi I, Gomportz R, Martin RL, Fox DJ, Keith T, Al-Laham MA, Peng CY, Nanayakkara A, Conzalez C, Challacombe M, Gill PMW, Johnson BG, Chen W, Wong MW, Andres JL, Head-Gordon M, Replogle ES, Pople JA (1998). Gaussian 98 (Revision A.7). Gaussian, Pittsburg

26. Casas JS, Castaño MV, Cifuentes MC, García-Monteagudo JC, Sánchez A, Sordo J, Abram U (2004) J Inorg Biochem 98:1009

27. Otto A, Mrozek I, Grabhorn H, Akemann W (1992) J Phys Condens Matter 4:1143

28. Dollish FR, Fateley WG, Bentley FF (1974) Characteristic raman frequencies of organic compounds. Wiley, New York, p 35

29. West DX, Carlson CS, Bouck KJ, Liberta AE (1991) Transit Met Chem 16:271

30. Beraldo H, Lima R, Teixeira LR, Moura AA, West DX (2001) J Mol Struct 559:99

31. Togni A, Hayashi T (eds) (1995) Ferrocenes: homogeneous catalysis, organic synthesis, materials science. VCH, Weinheim

32. Malaun M, Kowallick R, McDonagh AM, Marcaccio M, Paul RL, Asselberghs I, Clays K, Persoons A, Bildstein B, Fiorini C, Nunzi JM, Ward MD, McCleverty JA (2001) J Chem Soc Dalton Trans 2001:3025

33. Wong MW (1996) Chem Phys Lett 256:169