Supplementary Material (ESI) for Chemical Communications

Supplementary Information

Electrochemical evidences in oxidation of acetaminophen in the presence of glutathione and *N*-acetylcysteine

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1) General

Cyclic voltammetry and controlled-potential coulometry were performed using an Autolab model PGSTAT 20 potentiostat/galvanostat. The cell used was a simple and undivided cell. The working electrode used in the voltammetry experiment was a glassy carbon disk (1.8 mm diameter) and a platinum wire was used as the counter electrode. The working electrode potentials were measured versus SCE (all electrodes from AZAR Electrodes). The working electrode used in controlled-potential coulometry was an assembly of four carbon rods (6mmdiameter and 4 cmlength) and large platinum gauze constituted the counter electrode.

The homogeneous rate constants were estimated by analyzing the cyclic voltammetric responses using the DigiElchSB simulationsoftware.[1] An excellent fit between the experimental and simulated data was obtained over this range of experimental conditions for the following kinetic parameter values.

IR spectra were recorded on Perkin Elmer spectrum GX FT-IR spectrometer.

All reagents were reagent-grade materials from Aldrich. These chemicals were used without further purification.

2) Experimental Section

2.1) Electrochemical synthesis of glutathione disulfide (4) and diacetylcystine (5)

In a typical procedure, 80 ml of phosphate buffer in water (0.2 M, pH= 7.0) was pre-electrolyzed at 0.25 V versus SCE, in an undivided cell; then, 0.1 mmol of acetaminophen (**1**) and 2.0 mmols of glutathione (**3a**) or *N*-acetylcysteine (**3b**) were added to the cell. The electrolysis was terminated when the cathodic peak that corresponds to the reduction of *N*-acetyl-*p*-benzoquinone-imine (NAPQI) (**2**) (C_1 in Fig. 1) in cyclic voltammetry reappears. At the end of the electrolysis, the remained acetaminophen (**1**) was removed from the solution by extraction using chloroform. After decreasing the volume of solution to less than 10 ml (by evaporation) cell was placed in the refrigerator overnight. The precipitated solid was collected by filtration. After filtration: product **4** was characterized by a comparison of their FT-IR spectrum with that obtained previously.[2]

The electrochemical synthesis of **4** and **5** can also be performed by means of constant current electrolysis (ca. 1 mA cm^{-2}). Constant current synthesis was performed in same

conditions as described above. To take the high product yield and simple separation and purification, constant current synthesis can be perform in the absence of supporting electrolyte.

2.2) Simulation

A scheme for the electrochemical oxidation of acetaminophen (**1**) in the presence of glutathione (**3a**) and *N*-acetylcysteine (**3b**) is proposed tested by digital simulation. On the basis of an *EC′* mechanism, the observed homogeneous rate constants (k_{obs}) of reaction of NAPQI with glutathione (**3a**) and *N*-acetylcysteine (**3b**) have been estimated by comparison of the simulation results, (curves b, in Fig. 1-4, green curves), with experimental cyclic voltammograms (curves a, in Fig. 1-4, orange curves). The transfer coefficient (α) was assumed to be 0.5, and the formal potentials were obtained experimentally as the average of the two peak potentials observed in cyclic voltammetry. The heterogeneous rate constants are estimated by use of an experimental working curve.[3] The procedure is performed based on achieving the best fit between simulated and experimental cyclic voltammograms. The method is developed for a variety of scan rates and glutathione (**3a**) or *N*-acetylcysteine (**3b**) concentrations.

2.3) Experimental and simulated cyclic voltammograms of acetaminophen in the presence of glutathione

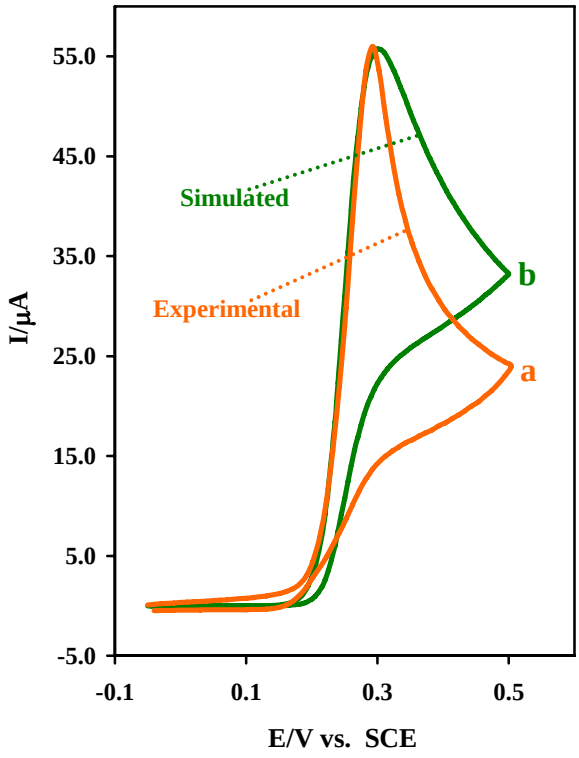


Fig. 1. Cyclic voltammograms of 1 mM acetaminophen (**1**) in the presence of glutathione (**3a**) (5 mM) (a) experimental (orange curve), (b) simulated (green curve). At a glassy carbon electrode, in phosphate buffer solution (c = 0.2 M, pH 7.0); scan rate: 100 mV s^{-1}; t = 25±1 °C.

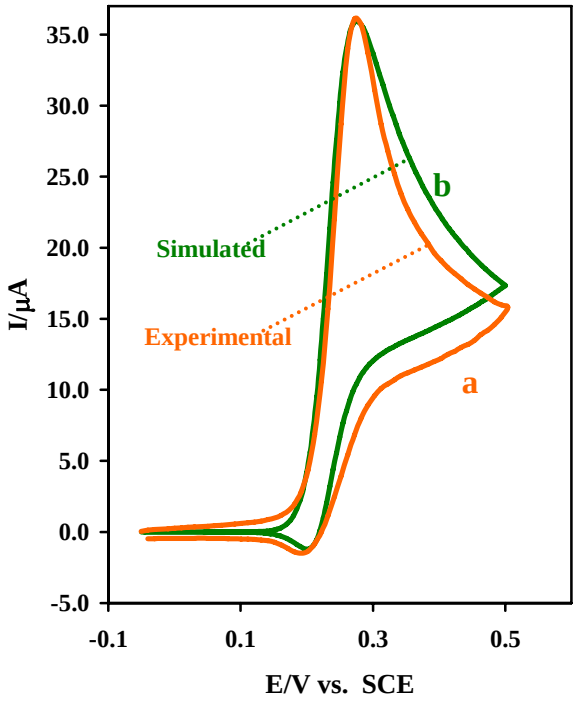


Fig. 2. Cyclic voltammograms of 1 mM acetaminophen (**1**) in the presence of glutathione (**3a**) (2 mM) (a) experimental (orange curve), (b) simulated (green curve). At a glassy carbon electrode, in phosphate buffer solution (c = 0.2 M, pH 7.0); scan rate: 100 mV s^{-1}; t = 25±1 °C.

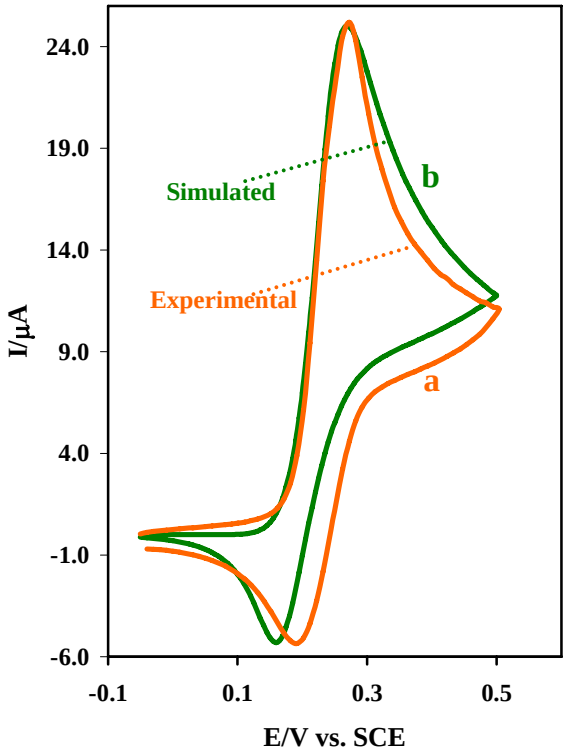


Fig. 3. Cyclic voltammograms of 1 mM acetaminophen (**1**) in the presence of glutathione (**3a**) (1 mM) (a) experimental (orange curve), (b) simulated (green curve). At a glassy carbon electrode, in phosphate buffer solution (c = 0.2 M, pH 7.0); scan rate: 100 mV s^{-1}; t = 25±1 °C.

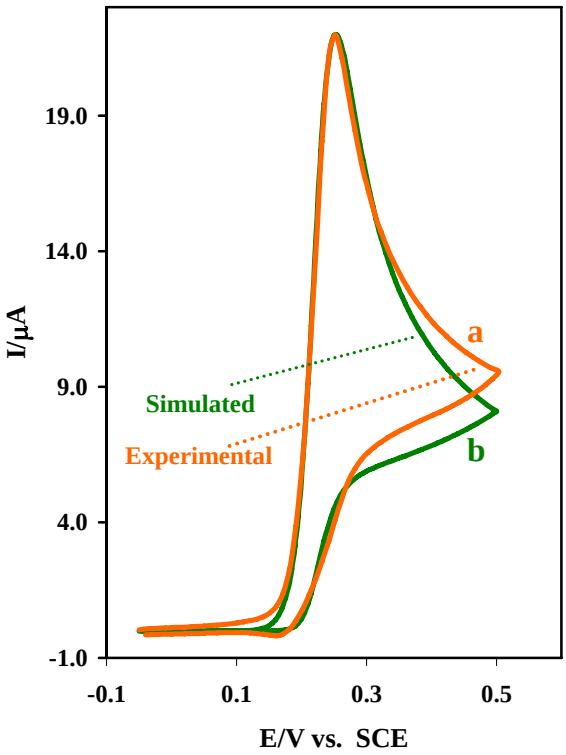
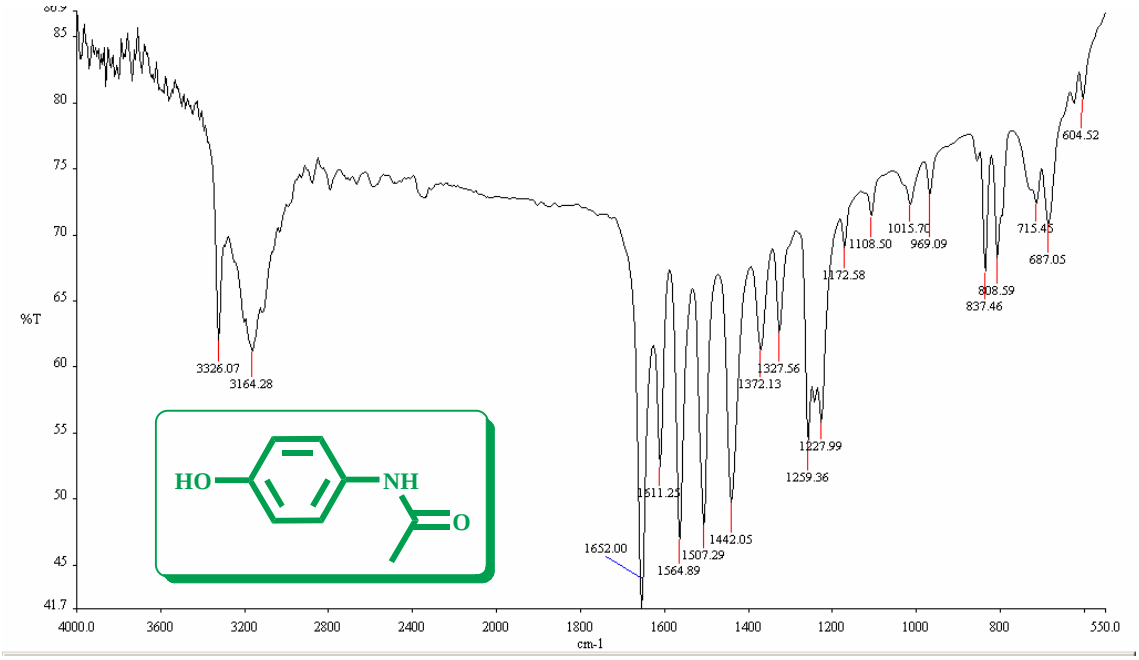


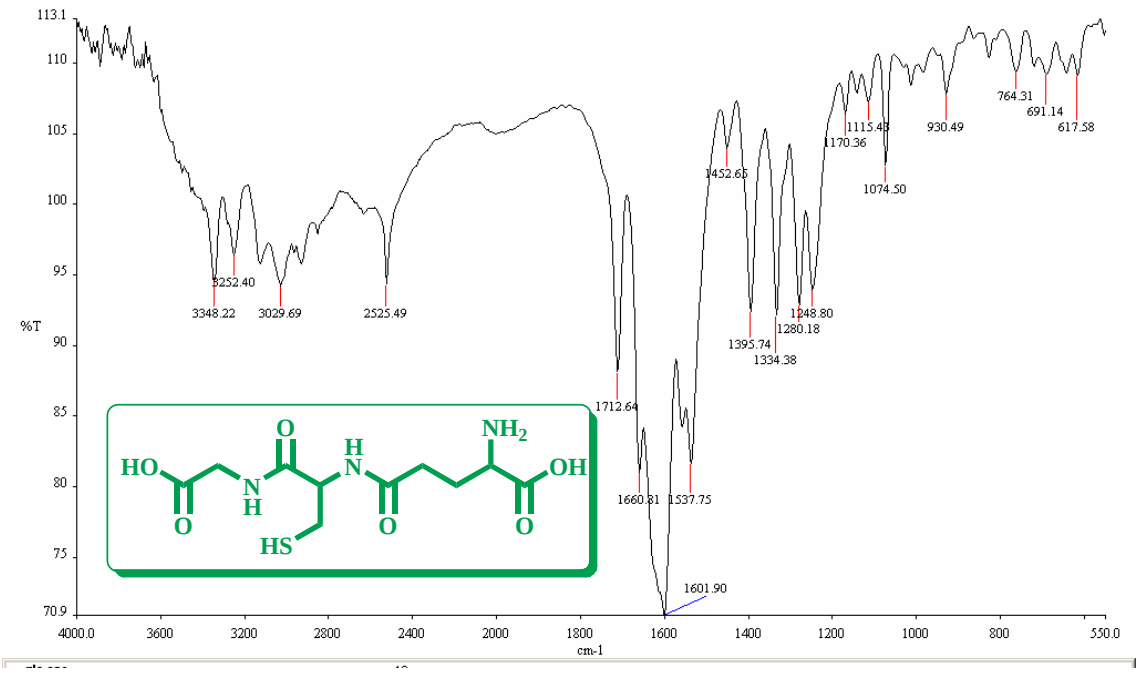
Fig. 4. Cyclic voltammograms of 1 mM acetaminophen (**1**) in the presence of *N*-acetylcysteine (**3b**) (1 mM) (a) experimental (orange curve), (b) simulated (green curve). At a glassy carbon electrode, in phosphate buffer solution (c = 0.2 M, pH 7.0); scan rate: 100 mV s^{-1}; t = 25±1 °C.

3) FT-IR Spectra

3.1) FT-IR spectrum of acetaminophen (1)

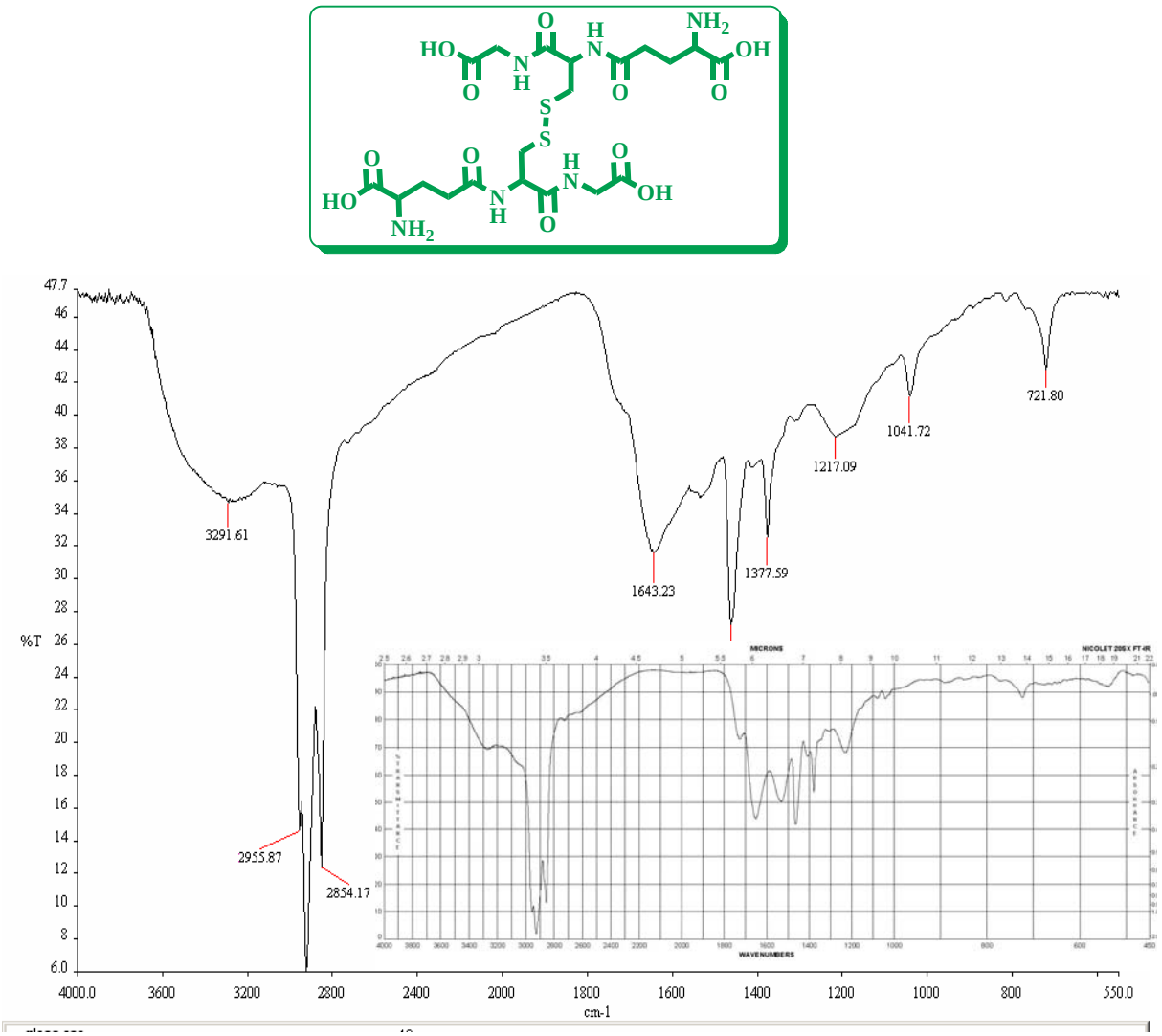


3.2) FT-IR spectrum of glutathione (3a)

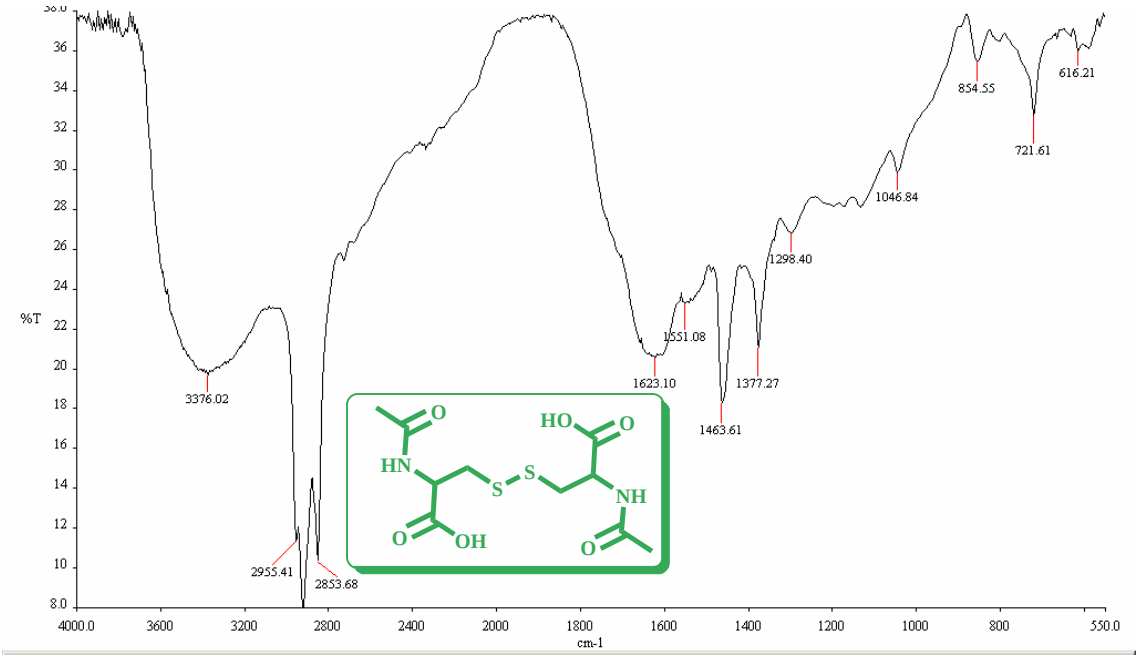


3.3) FT-IR spectrum (obtained from Nujol mull) of glutathione disulfide (4) (CAS number 27025-41-8) synthesized according to the present procedure.

Inset: FT-IR spectrum of glutathione disulfide obtained from "https://www.sigmaaldrich.com/technical-service-home/product-catalog.html"



3.4) FT-IR spectrum (obtained from Nujol mull) of diacetylcysteine (5) synthesized according to the present procedure.



4) References

1 M. Rudolph, *J. Electroanal. Chem.*, 2002, **529**, 97, See also: http://www.elchsoft.com/.

2 https://www.sigmaaldrich.com/technical-service-home/product-catalog.html .

3 R. Greef, R. Peat, L. M. Peter, D. Pletcher, J. Robinson, *Instrumental Methods in Electrochemistry*, Ellis Horwood, Chichester, UK, 1990, p. 189.